THE OHIO JOURNAL OF SCIENCE

VOL. LV SEPTEMBER, 1955 No. 5

CARBONIC ANHYDRASE IN CERTAIN SPECIES OF PLANTS

THOMAS E. BROWN AND H. CLYDE EYSTER

Charles F. Kettering Foundation, Yellow Springs, Ohio

The first evidence of carbonic anhydrase in leaves was obtained by Neish (1939), who observed activity in both the crude chloroplast and cytoplasm fractions of four species. Several investigators failed to find any evidence of carbonic anhydrase in leaves (Burr, 1936; Mommaerts, 1940; Roughton, 1934; Van Goor, 1948) both before and after Neish's observations were reported. Evidence for the general occurrence of carbonic anhydrase in leaves was presented by Bradfield (1947), with which all of the more recent work is in agreement. The following studies of plant carbonic anhydrase have recently been reported: distribution among different species (Bradfield, 1947; Day and Franklin, 1946; Osterlind, 1950; Sibly and Wood, 1951; Steeman et al., 1949; Waygood and Clendenning, 1950, 1951), intracellular localization (Day and Franklin, 1946; Waygood and Clendenning, 1950, 1951), pH and temperature relations (Day and Franklin, 1946; Sibly and Wood, 1951; Waygood and Clendenning, 1950, 1951), stability of leaf extracts (Bradfield, 1947; Day and Franklin, 1946; Sibly and Wood, 1951; Waygood and Clendenning, 1950, 1951), purification (Sibly and Wood, 1951; Sirois and Waygood, 1952; Wood and Sibly, 1952; Yen and Tang, 1951), zinc content (Sibly and Wood, 1951; Wood and Sibly, 1952), kinetics (Bradfield, 1947; Byerrum and Lucas, 1952; Steeman et al, 1949; Waygood and Clendenning, 1951; Wood and Sibly, 1952), including linked reactions with plant decarboxylases (Hansl and Waygood, 1952), effects of cysteine (Bradfield, 1947; Sibly and Wood, 1951; Waygood and Clendenning, 1950, 1951), effects of inhibitors (Day and Franklin, 1946; Sibly and Wood, 1951;Waygood and Clendenning, 1950, 1951; Wood and Sibly, 1952), and relative activity of extracts prepared from albino, etiolated, and normal leaf tissue (Waygood and Clendenning, 1950, 1951).

MATERIALS AND METHODS

Carbonic anhydrase catalyzes the hydration and dehydration of both carbonic acid and ammonium carbamate, involving both the uptake and output of carbon dioxide depending upon the pH of the medium. The reaction as used in the present work measured the carbon dioxide evolved from a given $NaHCO_3$ solution by contact with carbonic anhydrase contaniing plant macerates.

The method used in the present work was a modification of the boat method of Meldrum and Roughton (1933) which has served as the basic method of most workers to date. A standard, rectangular Warburg constant temperature bath (15°C) was fitted with an arm shaker driven by a $\frac{1}{5}$ h. p., 1750 rpm. DC motor with a chain drive and attached gear box to stabilize shaking speed. The shaking rate was controlled by a rheostat and measured with a tachometer.

The reaction vessel was a 50 ml. Erlenmeyer flask divided into two compartments by forcing the bottom upward with a knife edge to form a partition across the diameter of the vessel (Hove et al., 1940). A 2 cm. deep-wedged partition was found to provide two compartments which could hold up to 4 ml. each without accidental mixing and yet allowed complete mixing of the reactants upon shaking. Connections between vessel and manometer were combinations of glass and rubber pressure tubing. Two manometers were used; one a standard Warburg manometer from which the side arm had been removed (as used by Waygood and Clendenning, 1950) and the other a simple large-bore U-shaped manometer. The Warburg manometer was used in the majority of the activity measurements. The large-bore manometer was employed on the most highly active preparations.

All plant tissues whose carbonic anhydrase activity was to be determined, with the exception of the algae, were prepared by macerating 1.5 g. of fresh, healthy leaves with 3 g. banding sand (Central Scientific Co.) and either 5 ml. distilled deionized water or 5 ml. 0.01 M l-cysteine solution (as specified) in a small porcelain mortar using an oversize pestle. As an aid to maceration the leaves were cut into fine pieces with scissors before weighing. This technique provided a homogeneous sample after three minutes of grinding. The suspension was filtered with suction through 51 gauge nylon placed over a Gooch extraction funnel. Employing a water pump, filtration was rapid with all but the most viscous samples. The sand was retained almost entirely by the nylon. Determinations of enzyme activity were started immediately.

Two milliliters of 0.2 N solutions of c. p. $NaHCO_3$ made up in 0.02 N solution of c. p. NaOH were placed in one compartment of the vessel, and 2 ml. of 0.2 N phosphate buffer (pH 6.70) plus 1 ml. of plant tissue macerate were placed in the other. The reaction vessel was connected to the shaker arm and manometer and was then equilibrated for five minutes, a time period found adequate in preliminary tests. The contents of the two compartments were then mixed and readings were taken at 15 sec. intervals during continuous high speed shaking (200 oscillations per min., 6-inch thrust, 75 degree arc).

RESULTS AND DISCUSSION

Carbonic Anhydrase Activity of Different Plant Species

The carbonic anhydrase activity of several species and varieties is reported in figure 1. All species but New Zealand spinach are reported on here for the first time. New Zealand spinach was used as it was desired to include a species of known high activity for comparison (Waygood and Clendenning, 1950). With the exception of *Nostoc muscorum*, all plants were macerated in water. No activity could be found in *Nostoc* preparations except when macerated in 0.01 M cysteine, an ultrasonic generator being used to rupture the cells.

The question arose as to whether the physiological changes occurring in leaves preceding autumnal coloration would affect the carbonic anhydrase activity of such tissues as well. Mature green and senescent yellow leaves from the same branch of *Liriodendron tulipifera L.* were tested for their carbonic anhydrase activity. Carbonic anhydrase activity of the yellow leaves was about 55 percent lower than the activity for the green leaves. Autumnal leaf coloration in this species was thus accompanied by a considerable decrease in carbonic anhydrase activity.

With leaves of certain plant species the addition of cysteine to the grinding medium increased the carbonic anhydrase activity in agreement with Bradfield (1947), Waygood and Clendenning (1950) and Sibly and Wood (1951). An increase in activity of about 150 percent was noted on "low activity" plants when 0.01 M l-cysteine was used as the macerating medium. On "high activity" plants the increase was only 3-11 percent. These increases in activity were

corrected for the effect of the cysteine solution on the control. Addition of 60 mg. solid cysteine to the system to provide 0.1 M concentration immediately before equilibration did not result in a significant increase in enzyme activity although the rate of CO_2 production was slightly higher both in the presence and absence of macerated leaf tissue. Apparently the effect of cysteine will occur only when it is brought in contact with the plant material at the time the cells are ruptured. This effect appears to be one of stabilization of the enzyme during preparatory handling. That it may be due, in whole or in part, to limiting substrate seems reasonable but this study does not provide proof since the basis of specie comparison was equal amounts of plant macerate which did not lend itself to variation of enzyme concentration.

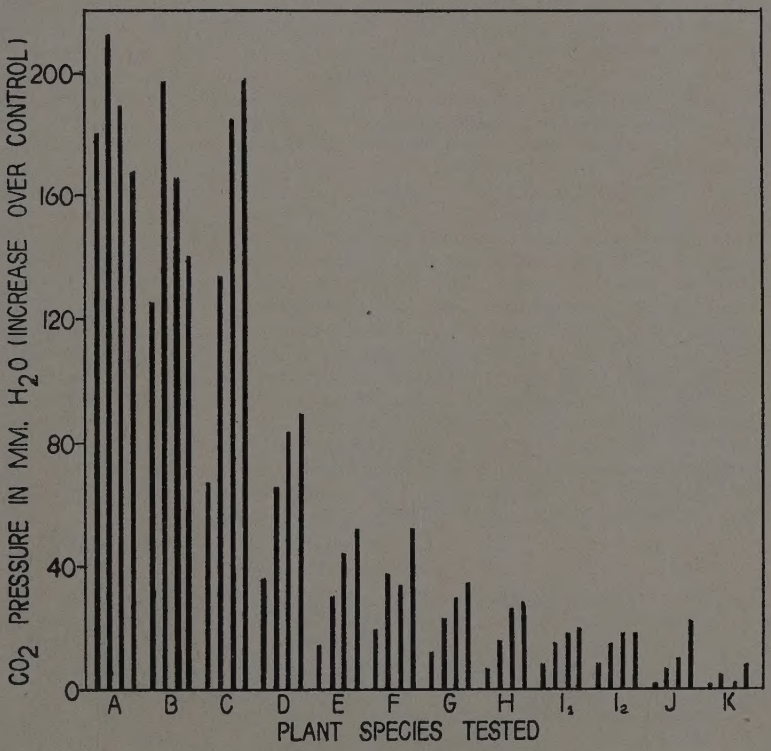


FIGURE 1. Carbonic Anhydrase Activity in Certain Plants (15, 30, 45, 60 sec. readings left to right in each group)

KEY:

A *Tetragonia expansa Murr.*
B *Ipomoea purpurea L.*
C *Amaranthus retroflexus L.*
D *Liriodendron tulipifera L.*
E *Maclura pomifera Schneid.*
F *Verbascum thapsus L.*
G *Nostoc muscorum Ag.*
H *Polystichum acrostichoides Schott*
I_1 *Zea mays L.*, green seedlings
I_2 *Zea mays L.*, albino seedlings
J *Plantago rugelii Dcne.*
K *Portulaca oleracea L.*

Carbonic anhydrase activity of different chloroplast pigment types of Zea mays L., and the effect of etiolation

Pedigreed strains of corn, segregating green and albino, green and yellow, and green, yellow and albino respectively, were tested for their carbonic anhydrase activity. Albino and "green" seedlings were grown from seed in both light and dark and pairs of samples were harvested at 10-14 days. All samples were macerated in 0.01 M cysteine to enhance the low carbonic anhydrase activity (figure 2).

The carbonic anhydrase activity of the light-grown plants belonging to four different pedigrees varied from a very low to a moderately high level of activity. The enzyme activities of albino plants from three different pedigrees were more uniform and were intermediate to the extremes observed on macerates of the green plants in the same pedigrees. The carbonic anhydrase activity of the green plants thus was higher or lower than that of the albino plants, depending upon the strain which was employed. No carbonic anhydrase activity could be found in macerates of either "green" or albino corn plants which were grown from seed in the dark. Two distinct pedigrees of yellow corn seedlings showed an even greater difference in carbonic anhydrase activity than was observed among the green plants in these pedigrees. Among the nine pedigrees of corn examined the highest and lowest enzyme activities were observed on the two yellow varieties which were grown in light. These findings present an interesting problem in genetics. The levels of carbonic anhydrase activity observed were a characteristic of the strain employed and were thus subject to genetic control. However, there is no simple relationship between the genetic control of pigmentation and carbonic anhydrase activity as observed on the leaf macerates *in vitro*.

Bradfield (1947) has reported that plants showing high carbonic anhydrase activity in their leaves do not show detectable activity in their roots. Waygood and Clendenning (1950) have observed much lower carbonic anhydrase activities in albino than in normal green barley leaf extracts as well as in the white regions of variegated *Tradescantia* leaves. Etiolation of *Tropaeolum* and *Petroselinum* leaves was also associated with a decrease in carbonic anhydrase activity (Waygood and Clendenning, 1950). Our observations on corn varieties are confirmatory with respect to the effect of etiolation. However, the large variation in levels of activity observed between green plants of different pedigrees and the yellow plants of different pedigrees suggests genetic control of enzyme activity which may bear no relation to chlorophyll content or photosynthetic capacity.

SUMMARY

Carbonic anhydrase activity was found in macerates of ten plant species (eight species of Angiosperms, *Polystichum acrostichoides* and *Nostoc muscorum*) which have not previously been examined for the presence of this enzyme. A decrease in carbonic anhydrase activity of leaves was found to accompany the decomposition of chlorophyll which is associated with autumnal leaf coloration in *Liriodendron tulipifera L.* Carbonic anhydrase activity was observed in macerates of genetically green and albino corn seedlings which were grown in light but no activity was observed in leaf macerates of corresponding plants which were cultivated in darkness. There was little relationship between pigmentation and level of carbonic anhydrase activity observed in extracts of nine varieties of *Zea mays L.* The levels of carbonic anhydrase activity were a varietal characteristic. An increase in activity of about 150 percent was noted on "low activity" plants when 0.01 M cysteine was used as the maceration medium. No increase in carbonic anhydrase activity was observed on addition of solid cysteine to leaf macerates in the reaction vessel immediately before equilibration. Apparently the effect of cysteine will occur only when brought in contact with the plant material at the time the cells are ruptured.

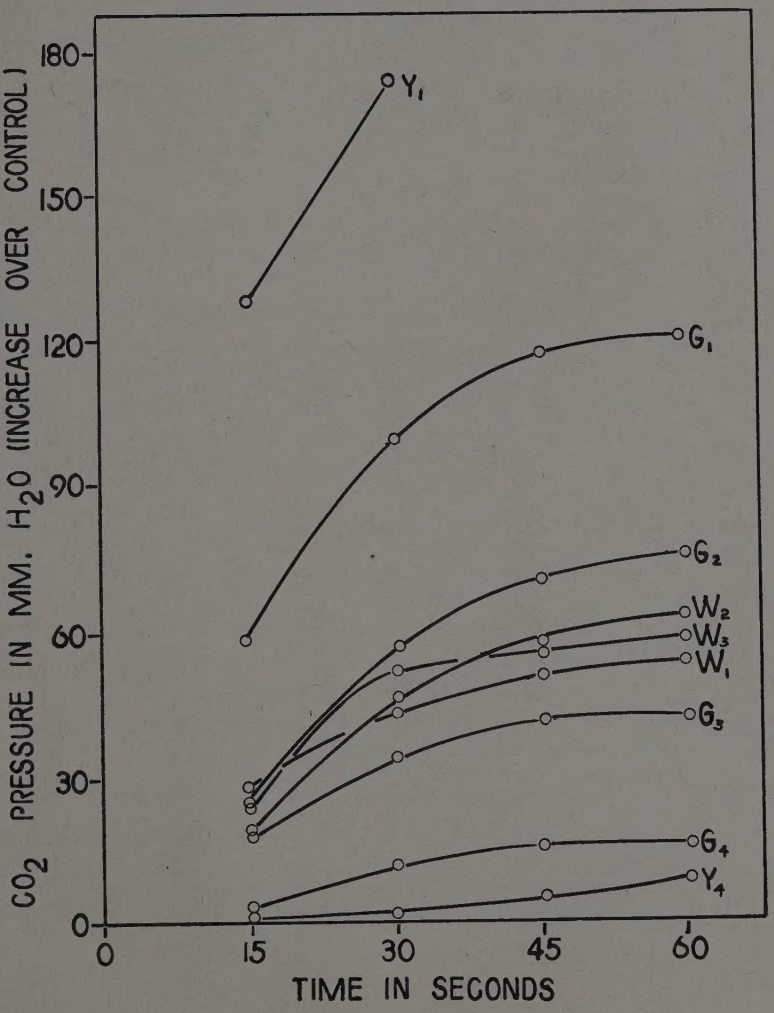


FIGURE 2. Carbonic Anhydrase Activity of Varieties of *Zea mays L.*

KEY:

Subscripts indicate pedigrees as follows: 1=4811; 2=482; 3=484; 4=485.

G. green seedlings
W. white seedlings (albino)
Y. yellow seedlings

ACKNOWLEDGMENTS

The authors wish to express their appreciation and thanks to Dr. Harry V. Knorr, who designed the special manometric equipment and supervised its construction. We also wish to thank Dr. K. A. Clendenning of the Kettering Foundation and Dr. B. S. Meyer of the Ohio State University for assistance in the preparation of the manuscript. This investigation was conducted during tenure by the senior author of a fellowship from the Charles F. Kettering Foundation, under the supervision of the junior author, Research Plant Physiologist with the Charles F. Kettering Foundation. The senior author currently holds a similar position with the Kettering Foundation.

LITERATURE CITED

Bradfield, J. R. G. 1947. Plant carbonic anhydrase, Nature 159: 467–468.
Burr, G. O. 1936. Carbonic anhydrase and photosynthesis. Proc. Roy. Soc. London B 120: 42–47.
Byerrum, R. U., and **E. H. Lucas.** 1952. Studies in plant carbonic anhydrase. II. Kinetic studies. Plant Physiol. 27: 111–120.
Day, R., and **J. Franklin.** 1946. Plant carbonic anhydrase. Science 104: 363–365.
Hansl, N., and **E. R. Waygood.** 1952. Kinetic studies of plant decarboxylases and carbonic anhydrase. Can. Jour. Botany 30: 306–317.
Hove, E., C. A. Elvehjem, and **E. B. Hart.** 1940. The relation of zinc to carbonic anhydrase. Jour. Biol. Chem. 136: 425–434.
Meldrum, N. U., and **F. J. W. Roughton.** 1933–1934. Carbonic anhydrase, its preparation and properties. Jour. Physiol. 80: 113–142.
Mommaerts, W. F. H. M. 1940. The possible occurrence of carbonic anhydrase in green leaves. Proc. Kon. Akad. Wetensch. Amsterdam 43: 1044–1049.
Neish, A. C. 1939. Studies on chloroplasts. II. Their chemical composition and the distribution of certain metabolites between the chloroplasts and the remainder of the leaf. Biochem. Jour. 33: 300–308.
Osterlind, S. 1950. Inorganic carbon sources of green algae II. Carbonic anhydrase in *Scenesdesmus quadricauda* and *Chlorella pyrenoidosa*. Physiol. Plantarum 3: 430–434.
Roughton, F. J. W. 1934. Carbonic anhydrase. Ergebn. Enzymfor. 3: 289–302.
Sibly, P. M., and **J. G. Wood.** 1951. The nature of carbonic anhydrase from plant sources. Aust. Jour. Sci. Res. B. 4(4): 500–510.
Sirois, J. C., and **E. R. Waygood.** 1952. Studies on the purification of plant carbonic anhydrase. Paper presented at A.I.B.S. Meeting before the Am. Soc. Plant Physiol., Cornell Univ., Ithaca, N. Y.
Steeman, N. E., E. Nielsen, and **J. Kristainsen.** 1949. Carbonic anhydrase in submerged autotrophic plants. Physiol. Plantarum. 2: 325–331.
Van Goor, H. 1948. Carbonic anhydrase, its properties, distribution, and significance for carbon dioxide transport. Enzymologia B. 13: 73–164.
Waygood, E. R., and **K. A. Clendenning.** 1950. Carbonic anhydrase in green plants. Can. Jour. Res. C. 28: 673–689.
———, and ——. 1951. Intracellular localization and distribution of carbonic anhydrase in plants. Science 113: 177–179.
Wood, J. G., and **P. M. Sibly.** 1952. Carbonic anhydrase activity in plants in relation to zinc content. Aust. Jour. Sci. Res. B. 5: 244–255.
Yen, Lung-Fei, and **P. S. Tang.** 1951. Experiments on the mechanism of Photosynthesis II. On spinach carbonic anhydrase. Chin. Jour. Physiol. 18: 43–50.

The Elements of Chromatography. *T. I. Williams.* Philosophical Library, New York and Blackie and Son, Ltd., London and Glasgow. 1953. 90 pp. $3.75.

This little book is very aptly titled. It appears to be an excellent introduction to the common methods of chromatographic analyses. It would be of especial interest to workers with no previous training in this field. The book commences with a historical review of chromatographic analyses, and is followed by chapters describing the methods in greatest use to the date of publication. These are: adsorption, partition, ion-exchange, and other miscellaneous forms. Other chapters deal with the treatment of colorless substances, the process of development, and the current uses of chromatography in industry.

JAMES N. LILES

DRAINAGE PATTERNS IN THE ALAMBRE VALLEY, ARIZONA

JOSEPH G. WARGO
Department of Geology, Miami University, Oxford, Ohio

The influence of the arrangement of platy elements present in some granitic rocks on the stream patterns that are developed upon these rocks has been noted in the Alambre Valley, in southern Arizona.

REGIONAL SETTING

The Alambre Valley is located between the Coyote and Quinlan Mountains in south central Arizona, approximately 30 mi. west of Tucson. The topographic appearance of this and adjacent areas has been outlined by Bryan (1922, 1923) and McGee (1897) and it need only be repeated here that the larger mountain masses exist as isolated peaks and ranges surrounded by alluvium which has accumulated, sometimes to great depths, in the broad outlying valleys. Within the mountainous areas, smaller, considerably less well developed valleys are found, of which the Alambre Valley is an example. The drainage developed in this valley provides a clue to the relationship between the stream patterns and the internal structures of the rocks that underlie the valley.

The portion of the Alambre Valley shown in figure 1 consists of approximately 3 sq. mi. of relatively flat land, enclosed on three sides by the rugged slopes of the Coyote and Quinlan Mountains. Alluvium in the valley is made up almost entirely of fragments derived from the weathering of the surrounding granodiorite, monzonite and pegmatite. That the alluvial cover is relatively thin is indicated by numerous small outcrops of bedrock some distance from the mountain front and by bedrock outcrops in the beds of shallow streams which flow in the central portion of the valley. Because of their size, these smaller outcrops have not been shown on figure 1.

RELATIONSHIP OF STREAM PATTERNS TO STRUCTURE

The dominant structural features of the area consist of: (1) the north-south trending Pan Tak Fault which outlines the western boundary of the Coyote Mountains and (2) the northwest trending, steeply dipping planar structures (foliation) developed in the rocks of both the Coyote and Quinlan Mountains (fig. 1). Within the granitic rocks of the Quinlan Mountains the planar structures are formed by preferred alignment of biotite flakes, feldspar porphyroblasts and hand-sized clots of a schistose, biotite-rich rock. The vast majority of the planar features strike approximately northwest and dip steeply to the southwest, giving the rock a distinct northwesterly "grain".

The master stream in the Alambre Valley is denoted the Pan Tak Wash on figure 1. Near the head of this stream, its channel follows the soft gouge zone of the Pan Tak Fault. Although outcrops of the Pan Tak Fault are not found farther down in the valley, a projection of the strike of the fault closely follows the present channel.

The initial step in the development of the stream channels involved the weathering of the foliated rocks. Weathering tends to preserve and accentuate the northwesterly "grain" of the rock and results in the formation of a series of small, rather rigidly oriented and poorly interconnected grooves that develop as the softer portions of the rock are removed. When the relief is slight, as on the bedrock floor of the Alambre Valley, the internal structure of the rock serves to guide the flow of water and stream channels are developed parallel to the north-

THE OHIO JOURNAL OF SCIENCE 55(5): 263, September, 1955.

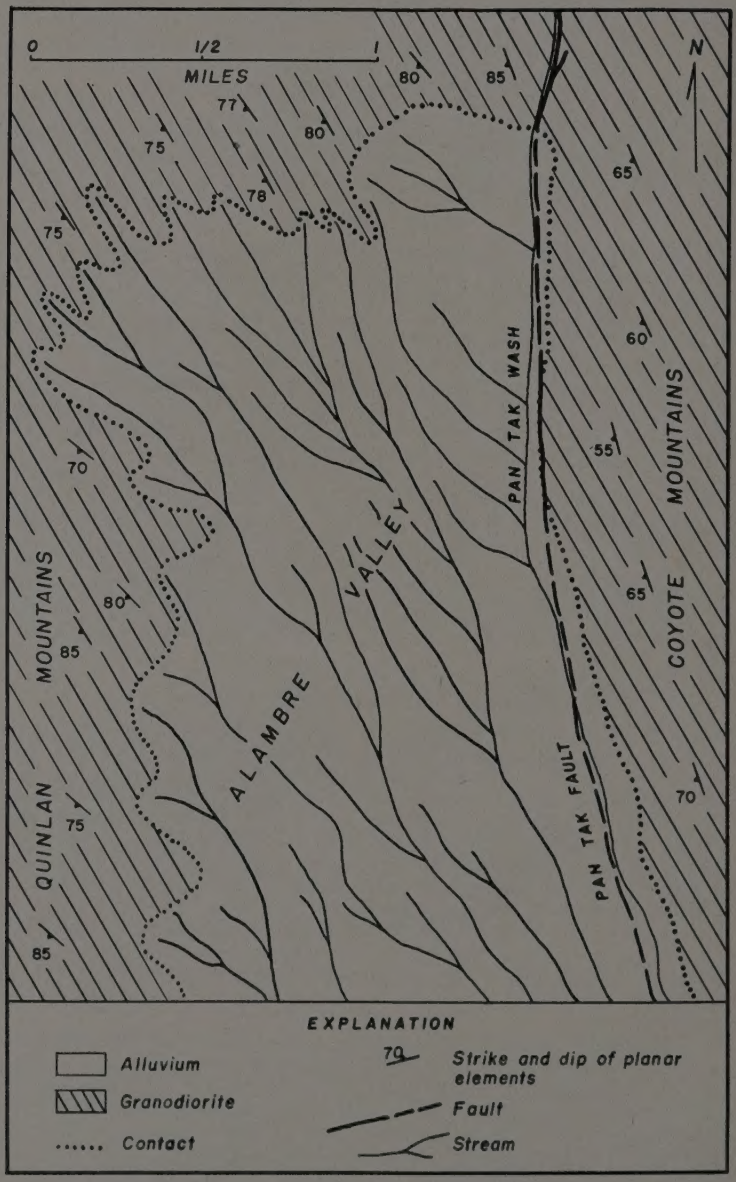


FIGURE 1. Geologic map of the Alambre Valley, Arizona.

westerly trend. In order to make an accurate comparison between the stream trends and the strikes of planar structures in the surrounding rocks, two distribution curves were plotted. The graph in figure 2 represents: (1) distribution of strike readings taken on planar structures in the Quinlan Mountains (solid line—135 readings) and (2) distribution of a number of directional readings taken

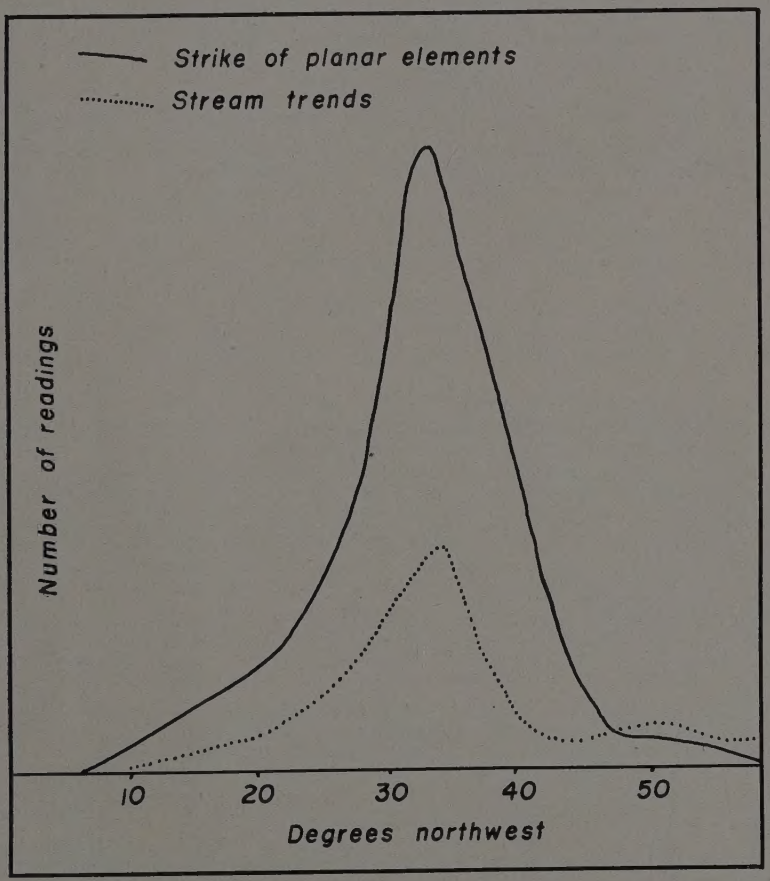


FIGURE 2. Distribution curves of stream trends and strike of planar structures, Alambre Valley, Arizona.

on short segments of streams in the Alambre Valley (dotted line—40 readings). The graph emphasizes the fact that the stream directions and the strikes of planar structures are essentially the same—approximately N34°W—and suggests that the development of the streams was directly influenced by the planar structures present in the bedrock floor of the valley.

The stream patterns developed in the Alambre Valley serve to illustrate that even the usually somewhat obscure planar features found in some plutonic rocks

influence topographic development on a scale at times comparable to that exerted by major faults and joint systems.

ACKNOWLEDGMENTS

This paper is an outgrowth of the study of internal structures in parts of the Coyote and Quinlan Mountains. The writer wishes to acknowledge the advice and assistance of Dr. R. L. DuBois of the University of Arizona. Mr. W. L. Kurtz read the manuscript and offered helpful suggestions.

LITERATURE CITED

Bryan, K. 1922. Erosion and sedimentation in the Papago country. U.S. Geol. Surv. Bull. 730-B.

———. 1923. The Papago country, Arizona. U.S. Geol. Surv. Water Supply Paper 499: 247.

McGee, W. J. 1897. Sheetflood erosion. Geol. Soc. Amer. Bull., 8: 112.

The Distribution and Abundance of Animals. *H. G. Andrewartha* and *L. C. Birch.* University of Chicago Press, Chicago. 1954. xv + 782 pp. $15.00.

This new tome is presented as a textbook of animal ecology. Actually it is an encyclopedic treatment of the autecology of terrestrial insects, especially those of economic importance. Within the limits set by the authors it is thorough, detailed, and it gives emphasis to the quantitative and statistical aspects of insect populations. Considerable stress is also given to the climatic and physiological phases of insect ecology. The main theme, expressed repeatedly throughout the book, is the "chance of the individual to survive and multiply." This is developed into a "unifying theory." Other approaches to the study of ecology are critized for lacking a "unifying theory."

Attention is almost always directed to individual species or individual insects. Occasionally reference is made to similar work on other invertebrates and rarely to related work on vertebrates. However, ecological relations of individual insects are treated exhaustively. The authors are critical in approach, but tend to labor their criticism of points of view other than their own.

Concrete examples of each point discussed are profusely supplied, drawn largely from British publications, especially the authors' own researches in Australia. The senior author is affiliated with the University of Adelaide; the junior author with the University of Sydney.

There are nearly 800 references, largely insect studies, and all are quoted in the test. There is a combination bibliography and author index. There are over 200 illustrations and all except four half-tone pictorials are charts and graphs, for the most part assembled from published literature. Each chapter ends with a good summary.

The task performed by these authors is enormous. They have brought together a vast amount of the pertinent information on the ecology of terrestrial insects. However, this reviewer is not convinced that they have accomplished their goal "to build a wide and satisfying general theory of ecology". After a very brief introduction to the field of animal ecology the text is narrowed to the consideration of the distribution and abundance of insects. This would make a appropriate title for the book.

As a textbook in the general field of animal ecology it would not be satisfactory. It would, however, be suitable for graduate students in entomology or those taking a specialized course in insect ecology. As a source book and guide for advanced students and professional entomologists concerned with ecological research on insects and economic problems of insect populations it is unsurpassed.

RALPH W. DEXTER

NOTES ON SOME SALAMANDERS OF WARNER'S HOLLOW, ASHTABULA COUNTY, OHIO

WILLIAM B. MUCHMORE

Department of Biology, University of Rochester, Rochester 3, New York

A few years ago there appeared in the pages of this journal an account of a biologically interesting spot in southwestern Ashtabula County, called Warner's Hollow (Joseph, 1950). This account indicates that Warner's Hollow is an especially fine habitat for salamanders, stating that "the Caudata are represented here by *Triturus viridescens* (most numerous), *Plethodon jordani*, and *Eurycea lucifuga*. These forms of the Caudata are so plentiful that they may be observed crawling about in the wooded areas and among the rocky ledges of the Hollow all day long." This report of the latter two species in northeastern Ohio is particularly noteworthy, inasmuch as *Eurycea lucifuga* has previously been recorded no farther north in Ohio than the southwestern section (Bishop, 1943) and *Plethodon jordani* is a species apparently confined to the southern Appalachian mountains (Bishop, 1943; Hairston, 1950).

Correspondence with Mr. Joseph, in an effort to clarify the above record has revealed that no specimens of the salamanders were collected and that the report was based upon identification by a third party. These facts cast grave doubt upon the validity of the record.

Nevertheless, when opportunities presented themselves in June and August of 1954, I visited Warner's Hollow to examine the salamander fauna at first hand. On the first occasion Mr. Joseph very kindly consented to guide me to the locations where, to the best of his recollection, the animals in question had been seen. Intensive searching by Mr. Joseph and me for seven hours on June 21st and by me for four hours on August 14th failed to turn up any specimens of either *Eurycea lucifuga* or *Plethodon jordani*. However, salamanders were found which might well have been mistaken for specimens of those two species.

In the general area where *Eurycea lucifuga* was purported to have been quite common, I collected a number of specimens of *Eurycea l. longicauda* (6 on June 21st, 12 on August 14th). Since no specimens of *E. lucifuga* were found, and since *E. l. longicauda* is certainly present in Warner's Hollow, it seems reasonable to conclude that the animals reported by Joseph were misidentified and were in reality *Eurycea l. longicauda*. Therefore, until more concrete evidence is at hand, *Eurycea lucifuga* must not be considered to range into northeastern Ohio.

The moisture-soaked debris beneath a dripping cliff was searched for specimens of *Plethodon jordani*, but without success. However, in this habitat was found a number of specimens of *Desmognathus o. ochrophaeus*, some of which were almost black in color. It is possible that similar dark specimens might in haste have been identified as *P. jordani* and that these were reported by Joseph. In any event, in the absence of specimens the report of *Plethodon jordani* in Ohio must be considered as erroneous.

Although not providing any species of great rarity, Warner's Hollow is indeed an excellent place for observation of a number of salamanders. In all, seven species were encountered on my two visits, and in all probability other species could be added in the cooler, damper months of the year. Species found are as follows:

1. *Notophthalmus viridescens viridescens* (Rafinesque).[1] Only four specimens of the Common Newt were found. They were all hidden under rocks, three in

[1]For a discussion of the proper name for this form see Smith (1953).

TABLE 1

Comparison of Specimens of Eurycea bislineata

	Specimens with Costal Grooves Numbering:			Dorsolateral Stripe on Tail Measuring:			Number of Vomerine Teeth:		
	14	15	16	<½ Tail Length	½Tail Length	>½ Tail Length	Range	Mean	Std. Dev.
E. b. bislineata..... (data from Mittleman, 1949)...	0	110 (53%)	100 (47%)	occasionally	usually	rarely	8–21	12.2	3.07
Specimens from Warner's Hollow.....	0	32 (80%)	8 (20%)	in none*	in 4* (13%)	in 26* (87%)	9–28	15.0±.56	3.54±.40
E. b. rivicola..... (data from Mittleman, 1949)...	100 (81%)	23 (18.2%)	1 (0.8%)	—	—	in most	8–27	15	3.73

*of 30 specimens with nearly complete tails

the stream bed and one at the top of the gorge. Joseph's statement that this species is "most numerous" may refer to the situation in another season or in a moister summer than this.

2. *Desmognathus fuscus fuscus* (Rafinesque). Several specimens of the Dusky Salamander were found in moist situations along the stream, usually under rocks at the very edge of the water. This species is far less common than the following.

3. *Desmognathus ochrophaeus ochrophaeus* Cope. The Allegheny Mountains Salamander was by far the most abundant salamander encountered in the hollow. It was found under rocks and debris in and along the stream bed and in spring banks. In June when the slopes of the gorge were still rather moist this species could also be found some distance from the stream in company with *Plethodon c. cinereus* and *Plethodon g. glutinosus.*

4. *Plethodon cinereus cinereus* (Green). The Red-backed Salamander was fairly common on the damp sides of the hollow and a few were found in the woods above the gorge in June. Fifteen specimens were collected, all of which were of the striped phase. One of these, a female (snout-vent length=42 mm.), was found with a cluster of 6 eggs under a deeply embedded flat rock at the top of the gorge. The eggs were in an early stage of development with the neural folds just closing, which would seem to indicate that they were laid 5 or 6 days previously. None of this species was found in August, when the woodland floor was considerably drier than it had been earlier.

5. *Plethodon glutinosus glutinosus* (Green). A number of specimens of the Slimy Salamander were found in fairly moist situations along the stream bed and low on the slopes of the gorge wall.

6. *Eurycea bislineata* ssp. The Two-lined Salamander is commonly found under rocks in the stream bed. Forty specimens were collected and several times that number were seen but not taken.

The subspecific status of these animals is uncertain. It is problematical whether they should be considered as *E. bislineata bislineata* (Green), or *E.bislineata rivicola* Mittleman, or intermediate between these two subspecies. In his description of *E. b. rivicola*, Mittleman (1949 p. 96) says, "the range boundary of *rivicola* and *bislineata* in northeastern Ohio and western Pennsylvania [the state line in his map, Plate VI] is artificial; whether the two races interdigitate or replace each other abruptly in this region remains to be determined." Also, he says (p. 94) that two specimens from Meadville, Pa. are "quite typical *bislineata.*" Since Warner's Hollow is situated only about 40 mi. west-southwest from Meadville, with the state line midway between the two locations, it is interesting to examine the present specimens with respect to the characters used by Mittleman to differentiate *E. b. rivicola* from *E. b. bislineata.* In table 1 measurements of the Warner's Hollow specimens are compared with the corresponding data for typical *E. b. bislineata* and *E. b. rivicola* as given by Mittleman (loc. cit.).

This tabulation of presumably diagnostic characters appears to indicate that the population in Warner's Hollow is closer to *E. b. bislineata* in respect to number of costal grooves, but closer to *E. b. rivicola* in respect to number of vomerine teeth and length of the dorsolateral stripe on the tail. Apparently then this population is intermediate in certain respects between the two forms. It seems possible, moreover, that in northeastern Ohio there is no sharp line of demarkation between the subspecies as described, but rather that there may be a fairly smooth gradation between the two, which, upon more critical analysis, might be resolved further into several clines of characters. The final solution to this problem can only be found by careful examination of extensive collections from a number of localities through Ohio and Pennsylvania.

7. *Eurycea longicauda longicauda* (Green). As mentioned above, 18 specimens of the Long-tailed Salamander were taken here. These were, without exception, found under rocks which were situated among coarse grass at the edge

of the stream. It would seem that in this locality the grassy habitat is preferred by this species since no specimens were found away from the grass even though similar conditions of rocky cover, moisture, and temperature exist at many other places in the Hollow.

The occurrence of *E. l. longicauda* in this locality emphasizes the fact that this species is very interesting from a biogeographical point of view. As Grobman (1944) points out, the northern boundary of the range of *E. l. longicauda* roughly parallels the southern limit of glaciation in the eastern U. S., but lies at some distance north of this limit (cf. Bishop, 1943, p. 422; Fenneman, 1938, p. 453). Apparently this species has been able, since the end of the last ice age, to extend its range northward along the Hudson, Susquehanna and Ohio river systems for varying distances. In most places this northward extension seems to have been much less than 100 mi., though in the broad Hudson River valley the species has been found at least 150 mi. north of the glacial drift border. Further, it appears that in only two recorded instances has the species been able to cross the divide into the Great Lakes drainage system. Bishop (1941) reports the occurrence of *E. l. longicauda* near Montour Falls, New York, a locality which is drained by Catharine Creek into Seneca Lake, thence via the Seneca and Oswego rivers into Lake Ontario. The present station in Warner's Hollow is along Phelps Creek, a tributary of the Grand River which flows into Lake Erie.

It may be of some significance that each of the divides here involved, i. e., the Seneca Lake—Chemung River and the Grand River—Mahoning River, has a rather gentle grade from the south and a maximum altitude only slightly in excess of 900 ft. Since this is so we might suspect that the species has entered the Great Lakes drainage across other divides which lie at about 900 ft. elevation or lower. Several such divides may be found in north central and northwestern Ohio and right across Indiana and Illinois; but we note further that most of these are 100 mi. or more from the glacial boundary, only one, namely that between Killbuck Creek and Black River in Medina County, Ohio, being less than 50 mi. north of the boundary. This latter region, which is similar to the Grand—Mahoning divide in respect both to elevation and to distance from the glacial boundary, might seem to be the only other really likely point of entrance of *E. l. longicauda* into the Great Lakes watershed. It seems pertinent to the discussion to point out here that the only known penetration of *Plethodon richmondi* into the Great Lakes drainage has been in this same region, i. e., into the Black River valley (Green and Walker 1954). Thus, we might with some confidence expect the occurrence of *P. richmondi* in Warner's Hollow as well as *E. l. longicauda* in the Black River valley.

The above discussion is admittedly based on very incomplete data. It should, however, demonstrate clearly that the biogeography of *E. l. longicauda* (and perhaps of other plethodontids) in Ohio and adjacent states would be an interesting and rewarding study.

REFERENCES

Bishop, S. C. 1941. The salamanders of New York. N. Y. State Mus. Bull. 324: 1–365.
———. 1943. Handbook of salamanders. Comstock Publishing Co. Ithaca.
Fenneman, N. M. 1938. Physiography of eastern United States. McGraw-Hill, New York.
Green, D. M., and **W. F. Walker.** 1954. A northern extension of the range of *Plethodon richmondi* in Ohio. Copeia 1954: 60.
Grobman, A. B. 1944. The distribution of the salamanders of the genus *Plethodon* in eastern United States and Canada. Ann. N. Y. Acad. Sci. 45: 261–316.
Hairston, N. G. 1950. Intergradation in Appalachian salamanders of the genus *Plethodon*. Copeia 1950: 262–273.
Joseph, J. M. 1950. A description of Warner's Hollow. Ohio Jour. Sci. 50: 134–135.
Mittleman, M. B. 1949. American caudata. VI. The races of *Eurycea bislineata*. Proc. Biol. Soc. Wash. 62: 89–96.
Smith, H. M. 1953. The generic name of the newts of eastern North America. Herpetologica 9: 95–99.

VARIATIONS IN POPULATIONS AND CELL DIMENSIONS OF PHYTOPLANKTON IN THE ISLAND REGION OF WESTERN LAKE ERIE[1]

WALTER JAMES HINTZ
The Franz Theodore Stone Institute of Hydrobiology, The Ohio State University, Put-in-Bay, Ohio

During the fall of 1952 and the winter of 1953 water samples were taken around the Island region of Western Lake Erie in an effort to determine the phytoplankton populations in that region. In the fall of 1953 and the winter of 1954 samples were taken at a single station to supplement the data of the previous year. Three-dimensional measurements of each species of organism were made, and the populations were calculated in volumetric units.

AREA STUDIED

Samples during the years 1952–53 were taken along a triangular course, having a perimeter of about 60 miles, north of South Bass Island in Western Lake Erie. All sampling during this period was done within the following: longitude 41° 40′N and 41°50′N; latitude 83°00′W and 81°25′W (fig. 1). During the fall of 1953 and the spring of 1954 all samples were taken from the State of Ohio Fish Hatchery dock in Fishery Bay of South Bass Island.

METHODS

The sampling methods employed were those described by Verduin (1951a). The samples were taken weekly at approximately 10-mile intervals. At each sampling stop a bucket was filled and one liter of this water was strained through a small disc of number 25 silk bolting cloth with an aperture size of .064 mm. The filtered material was then resuspended by washing in a measured amount of filtered water, usually 10 or 20 ml., depending upon the density of the sample.

The samples taken in this study were from the surface only. Studies by Chandler (1940) indicate that the vertical distribution of the phytoplankton is essentially uniform. The data from 42 depth series taken during 1949 also showed no consistent trend with depth (Verduin, 1951a).

The counting was done with a Sedgewick-Rafter cell and a Whipple ocular micrometer (Welch, 1948). When ten fields had been counted and an average number of organisms per field had been determined, the volumes of the organisms were calculated by making (under 430X magnification) usually ten measurements of each dimension. When the average cell dimensions were found, the cell volume was calculated on the basis of the shape of the cell, the shapes in most cases being spherical, cylindrical, or a rectangular box. Both the average number of organisms per mm.^{3} and the average volumes of their respective cells being known, it was possible to calculate the volume of phytoplankton per liter of lake water.

In the winter of 1954 it came to the writer's attention that some organisms might be escaping through the bolting cloth disc. To check this supposition 25 ml. of the filtrate was passed through an M. F. millipore filter (Lovell Chemical Co., Watertown, Mass.). This instrument, designed for bacteriological analysis of water, retains all particles greater than approximately 0.5 μ (Goldberg, 1952). The filtered material was counted and a significant loss of *Cyclotella melosiroides* (Kirsh.) Lemmerman from the bolting cloth disc was discovered. In subsequent collections a count was always made with the millipore filter, and the calculated volume was added to that obtained with the use of the bolting cloth. Organisms other than *C. melosirodies* occasionally passed through the bolting cloth, but the amounts were insignificant when total populations were considered.

[1]From a thesis presented in partial fulfillment of the degree Master of Science, The Ohio State University, 1954.

The counts and measurements were made with an accuracy of 85 percent or better. Whipple (1927) states that when the Sedgewick-Rafter method is employed, two examinations of the same sample seldom differ by more than 10 percent. Serfling (1949) also found this to be true. Burkholder (1929) in his studies of Lake Erie found his error in counting diatoms to be about 18 percent. It is possible that an error may be introduced in the volumetric estimations as the phytoplankton cells are not always perfect cylinders, spheres, or rectangular boxes. The error attributable to this factor, however, is small; *i. e.*, if one were to assume that a rectangular box 10 μ x 10 μ x 100 μ were a cylinder 10 μ x 100 μ the volume obtained from this approximation would be about 22 percent too small. The geometric figures usually represent a better fit than in this hypothetical example.

The following references were used in identifying the organisms: Pestalozzi (1942), Smith (1933), Needham (1927), and Tiffany and Britton (1952).

RESULTS AND DISCUSSION

The bulk of the phytoplankton populations in Western Lake Erie in the fall, winter, and spring seasons are members of the Bacillariophyceae. The writer has no data showing what the major components were during the summer months; however, studies by Chandler (1940) show an increase in green and blue-green algae during this period.

The results of this study are presented as two separate groups of data: those samples taken over a wide range of territory and those taken at a single station.

The major components during the years 1952–53 are as follows: *Melosira* sp., *Asterionella formosa*, *Fragellaria* sp., *Cyclotella melosiroides*, *Tabellaria floculosa*, and *T. fenestrata* var *asterionelloides*. In the early fall and late spring, a green alga, *Pediastrum*, also appeared in significant numbers (averaging more than 1 percent of the populations during the fall months). Those found in insignificant numbers included *Aphanizomenon* sp., *Binuclearia* sp., *Anabeana* sp., *Polycystis* sp., and *Stephanodiscus* sp.

Two main types of communities were dominant throughout the year. *Asterionella-Melosira* made up the bulk of the populations during the fall and winter of 1952. In the spring of 1953, *Tabellaria* replaced *Melosira* as a major component, and there was also a significant amount of *Cyclotella*. During this season, *Tabellaria* made up approximately 42 percent of the total populations while *Asterionella* accounted for about 22 percent and *Cyclotella* and *Melosira* each made about 17 percent of the total.

Asterionella formosa Hass. was found as a co-dominant during all three seasons. On a yearly basis, therefore, it should be considered the major component. It is apparent that this diatom thrives under a wide range of physical conditions. Lund (1938) found it to have a temperature range of about 1.5° to 24°C.

It was stated earlier that significant amounts of *Cyclotella melosiroides* escaped through the bolting cloth. In the spring of 1954, it was found that from 45 percent to 79 percent of the total *C. melosiroides* population escaped. Since the cell size of this species was about the same in 1953 as in 1954 similar amounts were probably being lost that year. It must be kept in mind, therefore, that all stated figures of *Cyclotella* populations which were computed prior to March of 1954 may be considerably below the correct amounts.

Figure 1 shows average total populations during the fall sampling trips of 1952. The numbers shown are $\mu^3/l \times 10^{-8}$ and are averaged from samples taken within an area of approximately 100 square miles. *Melosira* made up from 80 to 90 percent of the total populations with *Asterionella* and *Pediastrum* making up the difference. Figure 1 shows the relative positions of these populations, the largest being found to the north and east of Pelee Island. A similar map of the spring, 1953, collections indicated relatively uniform populations throughout the island area.

Wright and Tidd (1933), who made a limnological investigation of Western Lake Erie in 1929–30, reported large and highly consistent inequalities in horizontal distribution of phytoplankton in Western Lake Erie as a whole. Their area was much larger, including Maumee Bay, and the waters near the mouth of the Detroit River.

Qualitatively speaking it was found that, from the standpoint of horizontal distribution, the differences in species composition were negligible. The types of phytoplankton taken at a single station in 1953–54 may therefore be considered representative of the species occurring at the other stations during the same period.

During 1953, the fall community paralleled that of 1952 as to quality; *Melosira* making up some 95 percent of the total populations until late December. In the winter and spring of 1954, however, *Cyclotella* was much more abundant making up from 75 percent to 95 percent of the total populations. The single

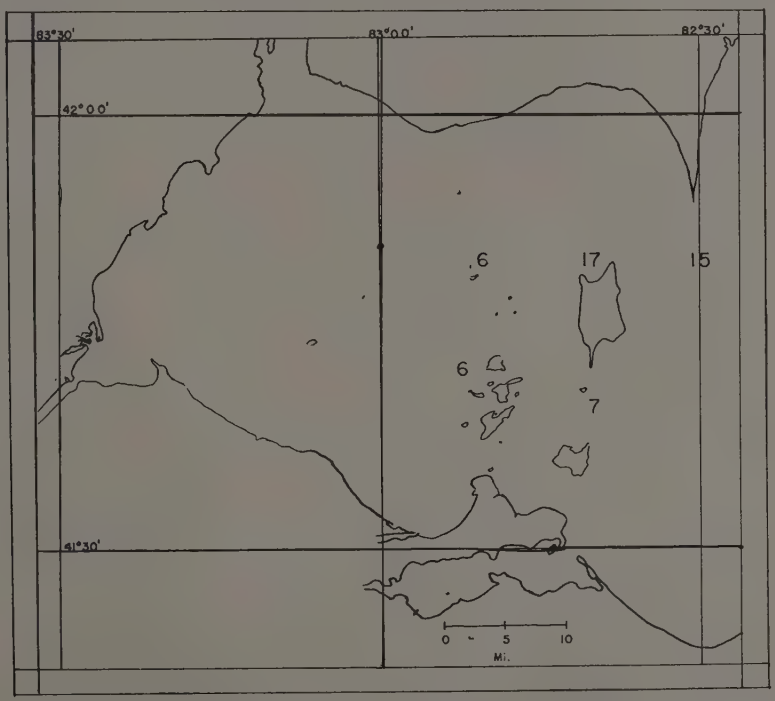


FIGURE 1. Map of Western Lake Erie showing horizontal distribution of phytoplankton populations during the fall of 1952. P identifies Pelee Island. Numbers represent μ^3 per liter ($\times 10^{-8}$).

station data for the two-year period are shown in figure 2. The population peak in 1952–53 came somewhat earlier than that in 1953–54; in the former season the maximal populations were found during January and February, while in the latter season the maximum was reached in April. The fact that the 1953–54 figures of *Cyclotella* populations were so much larger may be attributed to the introduction of the millipore filter in March of 1954.

A comparison of the populations calculated in this study and those found by Chandler (1940, 1942, 1944), Verduin (1951a, 1951b), and Curl (1950) is presented in table 1. Such comparison of data requires a number of explanations:

TABLE 1

Comparisons of Phytoplankton Data

Genus	1938-39[a]	1940[a]	1941[a]	1942[a]	1949[b]	1950	1951[c]	1952–53	1953–54
Asterionella	13.0 (Mar.)	9.4 (May)	70.0 (April)	32.8 (Mar.)	6.0 (April)	35.0[b] (May)	1.7 (Mar.)	6.5 (Feb.)	4.9 (Feb.)
Tabellaria	1.9 (Mar.)	8.4 (May)	2.1 (April)	59.1 (April)	200.0 (April)	NR	3.5 (May)	15.2 (May)	9.3 (Mar.)
Melosira	17.0 (Sept.	71.4 (Nov.)	32.9 (Oct.	59.1 (Sept.	NR	109.5[c] (Oct.)	1.80 (May)	41.0 (Sept.	9.7 (Dec.)
Cyclotella Melosiroides	NR	NR	NR	NR	NR	35.0[b] (May)	.75 (April)	40.5 (Feb.)	90.0 (April)
Stephanodiscus	50.2 (Oct.)	6.8 (Sept.)	2.8 (Sept.-Dec.)	2.8 (Sept.)	35.0 (May)	NR	NR	12.7 (Mar.)	.38 (Oct.)

Month maximum occurred is in parentheses
All values are x $10^6\mu^3$/liter
NR: not reported
a: data of Chandler (1938–1942) (reported in μ^3 as determined by this study)
b: data of Verduin (1949)
c: data of Curl (unpubl. MS Thesis, Ohio State Univ.)

1) all data of Chandler have been converted to units described in this paper on the basis of average cell sizes as determined here. Chandler arbitrarily accounted for variations in cell sizes by enumerating plankters as units per liter, the number of cells in a unit being dependent upon the size of the species. Thus eight cells of *Asterionella* or eight cells of *Tabellaria* were given equal weight with one cell of *Stephanodiscus*. The volume of one cell of *Asterionella*, however, was found, in the present study, to approximate 1000 μ^3, while one cell of *Stephanodiscus* varied between 16,000 and 20,000 μ^3. Eight cells of *Tabellaria*, on the other hand, were found to have a volume of from 20,000 to 30,000 μ^3. Conversion of Chandler's data, therefore, brought higher values than those indicated on his graphs. 2) Chandler found a species of *Cyclotella* similar in size to *Stephanodiscus*,

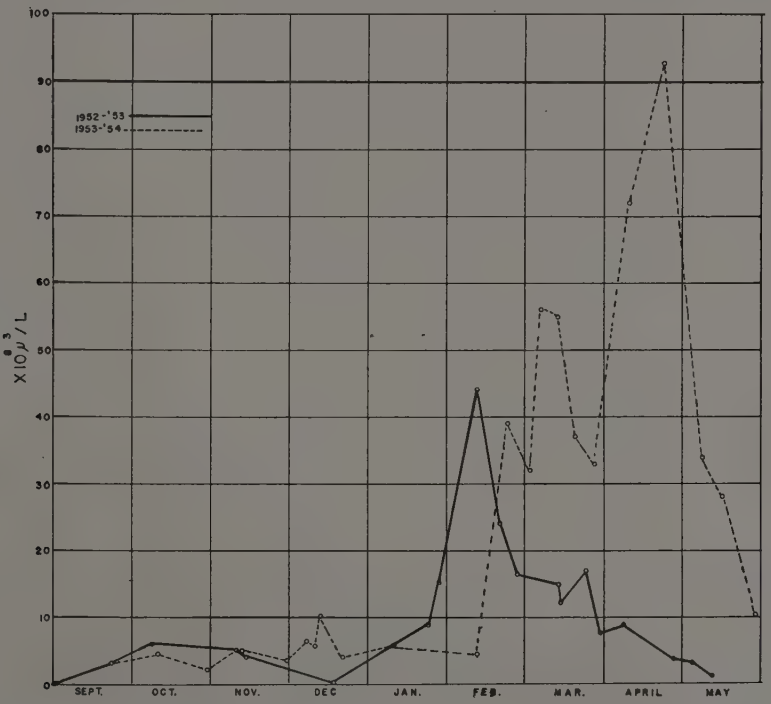


FIGURE 2. Graph showing phytoplankton populations in the fall, winter, and spring of 1952–54. Single station data.

while the data of Verduin, Curl, and the writer includes large populations of *Cyclotella melosiroides*. The appearance of this species corresponds with a drop in populations of *Stephanodiscus*. No records of *C. Melosiroides* were found prior to 1950. 3) Chandler reports his populations in maxima and minima; thus the maxima of comparable studies are given. It should also be borne in mind that Chandler's figures are from a single station. 4) Two species of *Tabellaria* are found abundantly in Western Lake Erie: *T. floculosa* and *T. fenestrata* var *asterionelloides*. Chandler found the former to make up the bulk of his *Tabellaria* populations. The writer, on the other hand, found the latter in greater numbers than did Chandler. In both studies the two species were combined for ease of computation.

Examination of table 1 shows a wide range of values. Conversion of Chandler's *Melosira* data was difficult because of great variations in cell size. A strand of *Melosira* 100 μ long varies from 2800 to 8000 μ^3. Examination of the month's data during which the maximum was reached indicates that the period in which the peak population of the individual species was reached has not radically changed. The *Melosira* peak appearing in December of 1953–54 may have been sub-maximal if samples had been taken in August and early September when *Melosira* is usually dominant.

Table 1 shows large variations in the maxima from year to year, and shows the conspicuous absence of *C. melosiroides* before 1950. The maxima of *Asterionella*, *Tabellaria*, and *C. melosiroides* occurred in spring, and the *Melosira* maxima usually occurred in the fall.

One objective of this study was to investigate possible correlations in variations in cell dimensions within certain species, with seasonal and horizontal distribution. Vegetative reproduction of diatoms involves formation of valves within the mother valve. These daughter valves produce new frustules, etc., throughout the vegetative growth period (Boyer, 1926). Because new frustules are formed within the elder a constant diminution should take place, at least in forms where expansion cannot take place. Such diminution is not true in some forms, however (Fritsch, 1935). Reduction is presumed gradual, therefore, until an auxospore is formed and an enlarged cell is produced. Diminution in size might theoretically continue indefinitely were there not formation of rejuvenescent cells of much larger size (Smith, 1933). Testing the hypothesis of cell diminution is difficult when whole populations are taken into consideration. Repeated auxospore formations would keep the cells from attaining the minimal size. Influx of new populations into the sampling area or sampling in different communities would also affect a curve of cell sizes.

An attempt was made in this study to determine whether diminution actually occurred among the major components of the diatom populations. The graduations of the ocular micrometer used in measuring were equal to 3.3 μ.

Average cell dimensions were tabulated for all of the major components, the dimension used being the diameter in those belonging to the Centrales (*Melosira* and *Cyclotella*) and the width in members of the Pennales (*Asterionella* and *Tabellaria*). That the other dimensions would be affected through vegetative reproduction is unlikely.

Two separate populations of *Melosira* were considered; one with a diameter of between 10 and 12 μ and one with a diameter between 5 and 7 μ. Examination of a curve of average diameters seems to indicate a decrease as the fall months progress and an increase from March through May. Apparently no diminution occurred at this time or if it did, it may have been offset by auxospore formation or by the influx of a new population. No auxospores were observed in the samples, however, and observation at other times seems to indicate auxospore formation during the summer months.

Cells of *Asterionella formosa* were found to have a fairly constant diameter throughout the year with variations between three and four micra. Samples taken at a single station during the period when ice cover made mobile sampling impossible did not indicate a significant trend in this dimension.

Widths of *Tabellaria* cells were found to differ by almost two micra over a period of three months, the median diameter being approximately 8.8 μ. Early spring measurements indicated a fairly rapid decline from 9.3 to 7.7 μ over a period of two weeks. If this decline was due to diminution through vegetative reproduction then the vegetative existence of the individual cells was short. A slight rise was indicated in April with another rapid drop in May.

Of the species studied, *Cyclotella* was found to have the greatest variation in cell diameter. A decline of from 10.6 to 7.9 μ in 14 days was observed. Though

the magnitude is greater the time of decline paralleled that of *Tabellaria*. Because *Cyclotella* was found to be particularly abundant in the winter months of 1953, the diameters of cells collected at a single station during the winter and spring of 1953 and 1954 were averaged and graphed (fig. 3). The 1953 data show a slow increase of one micron during the winter and a decline from 12.5 to 7.9 μ over a period of over two months. Such slow decline might be considered due to vegetative reproduction. The 1954 data, on the other hand, show a gradual increase in cell dimensions. This increase in cell size was noted in the fraction of *Cyclotella* recovered with the millipore filter over this same period. The trend

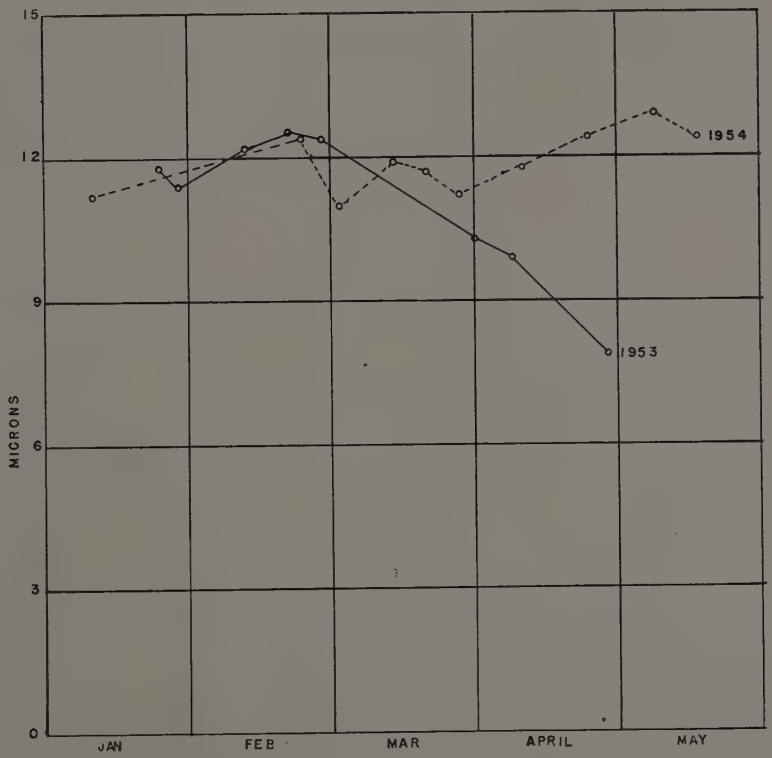


FIGURE 3. Average diameters of *Cyclotella melosiroides* graphed against time.

of cell dimension changes, therefore, were inconsistent and did not provide evidence supporting the theory of cell diminution during vegetative growth. The fact that auxospores were never observed in the collections makes it unlikely that repeated auxospore formation in the community obscured actual trends.

Studies of cell dimensions with reference to the horizontal distribution of the species showed no appreciable differences relative to position in the lake. Though the size of *Cyclotella* cells varied on the average by almost one micron in some areas, no significant trend of horizontal distribution was indicated. The dimensions of the other species usually did not vary by more than 0.4 μ.

SUMMARY

Phytoplankton studies showed average crops of 6 to 17 x 10^8 μ^3/l during 1952–53 throughout the island region. Single station data during the same seasons in 1953–54 showed higher crops largely due to the introduction of a new filtering device, the M. F. millipore filter, which retained quantities of *Cyclotella melosiroides* that were lost by bolting cloth filters.

Single station data during the years 1952–53 indicated the peak of the population occurred in February while in 1953–54 two pulses occurred; one in March and one in April.

Melosira comprised more than 90 percent of the fall phytoplankton volume during the two fall periods studied.

A diatom unrecorded for Western Lake Erie prior to 1950, *Cyclotella melosiroides*, has become a major component of the phytoplankton populations. 75 to 95 percent of the phytoplankton volume in the winter and spring of 1954 was composed of *Cyclotella melosiroides*.

Graphs of variations in cell dimensions of certain diatoms show a variety of trends not consistent with the theory of gradual diminution of vegetative cell size and subsequent increase due to auxospore formation. *Asterionella formosa* was found to have a fairly constant cell width throughout the season in contrast to *Cyclotella melosiroides* which varied from approximately 7 to 14 μ.

ACKNOWLEDGEMENTS

This study was conducted at the Franz Theodore Stone Institute of Hydrobiology at Put-in-Bay, Ohio. The writer wishes to acknowledge the valuable aid given by Dr. Jacob Verduin, who served as adviser, and by Dr. Thomas H. Langlois, Director of the Institute.

LITERATURE CITED

Boyer, C. S. 1926. Synopsis of North American Diatomaceae. Pt. 1. Proc. Acad. Nat. Sci., Philadelphia 68.

Burkholder, P. R. 1929. Microplankton studies of Lake Erie. Bull. Buffalo Soc. Nat. Hist. 14: 73–93.

Chandler, D. C. 1940. Limnological studies of Western Lake Erie. I. Plankton and certain physical-chemical data of the Bass Islands region, from Sept. 1938 to Nov. 1939. Ohio Jour. Sci. 40: 291–336.

———. 1942. III. Phytoplankton and physical-chemical data from Nov. 1939 to Nov. 1940. Ohio Jour. Sci. 42: 24–44.

———. 1944. IV. Relation of limnological and climatic factors to the phytoplankton of 1941. Trans. Amer. Microsc. Soc. 63: 203–236.

Curl, H. C. 1950. The distribution of phosphorus in Western Lake Erie and its utilization by natural phytoplankton populations. M. Sc. Thesis. The Ohio State University. Columbus.

Fritsch, F. E. 1935. The structure and reproduction of the algae. Cambridge University Press, London.

Goldberg, E. D., M. Baker, and **D. Fox.** 1952. Marine sampling with the molecular filter. Sears Found. Jour. Mar. Res. 2: 194–204.

Lund, J. W. G. 1938. Studies on Asterionella Formosa Hass. Jour. Ecol. 38: 1–35.

Needham, J. G., and **P. R. Needham.** 1927. A guide to the Study of Fresh Water Biology. The American Viewpoint Society, Inc., New York.

Pestalozzi, G. H. 1942. Das Phytoplankton des Susswassers. Die Binnengewasser. 16: 367–589. Diatomeen 2nd half.

Serfling, R. E. 1949. A Quantitative estimation of plankton from small samples of Sedgewick-Rafter cell mounts of concentrated samples. Trans. Amer. Microsc. Soc. 68: 185–198.

Smith, G. M. 1933. The Fresh-Water Algae of the United States. McGraw-Hill Co., New York.

Tiffany, L. H., and **M. Britton.** 1952. The Algae of Illinois. The University of Chicago Press, Chicago.

Verduin, J. 1951a. A comparison of phytoplankton data obtained by a mobile sampling method and those obtained from a single station. Amer. Jour. Bot. 38: 5–11.

———. 1951b. Comparison of spring diatom crops of Western Lake Erie in 1949 and 1950. Ecology 32: 662–668.

Welch, P. S. 1948. Limnological Methods. The Blakiston Co., Philadelphia.

Whipple, G. C. 1927. Revised by G. M. Fair and M. C. Whipple. Microscopy of Drinking Water. John Wiley and Sons, New York.

Wright, S., and **W. Tidd.** 1933. Summary of Limnological Investigations of Western Lake Erie in 1929 and 1930. Trans. Amer. Fish. Soc. 63: 271–285.

A REVIEW OF THE TRACHYLOEINI OF AMERICA NORTH OF MEXICO (*COLEOPTERA, CURCULIONIDAE*)

ELBERT L. SLEEPER

Department of Zoology and Entomology, The Ohio State University, Columbus 10

This paper is the result of a study started originally on the genus *Cercopeus* Schönherr. However, to establish the proper relationship of the related genera it became necessary to review all the genera and species in the tribe. The author hopes that this review will enable other workers to determine their material in this group which heretofore has been rather neglected. The author feels that further study should be made in Western United States for more members of this tribe.

DISCUSSION

The tribe Trachyphloeini is typified by the absence of ocular lobes, the eyes always free and rounded; the scrobes are variable, never at the same time linear and directed beneath; the outer segments of funicle moniliform; elytra scarcely surpassing the width of the prothorax at base; humeral angles rounded; metasternum short; the intercoxal lobe on the first abdominal segment large and broad; the corbels of the posterior tibiae open and the tarsal claws free.

All members of this tribe known to science are apterous. The larvae of all species breed in the roots of herbaceous plants and trees. They are potentially of economic importance, particularly in areas which have been recently cleared for cultivation of agricultural crops.

Occasionally the adults will be found feeding on foliage during the day, but with few exceptions they are night feeders, spending the day in leaf litter and rubbish around the host plant.

All of the species probably over-winter as an imago. The author has found sifting during the winter to be the most successful method of obtaining examples in Eastern United States, particularly those members of the genus *Cercopeus* Schönherr.

Males are very rare in this tribe. More than 500 weevils belonging to this tribe were examined and of this number only 2 were males. For this reason all descriptions, with the exception of *Pseudocercopeus setosus* n. gen. and n. sp., were drawn from the female. The description was not taken from the female of the afore mentioned because most of the appendages were lacking.

The type of *Cercopeus chrysorrhoeus* (Say) has been lost. A neoholotype has been designated. There might be some doubt as to which species of *Cercopeus* Say had. So far as the author is able to ascertain there are only two species of *Cercopeus* occuring in Pennsylvania, the type locality of *chrysorrhoeus*. They are the species I have designated as *chrysorrhoeus* and *maspavancus* n. sp. The first fits the original description very well in size, color pattern and general form, while *maspavancus* is much larger and differs from the original description in many ways. Also, though irrelevant, the one designated as *chrysorrhoeus* is the one most commonly accepted as that in collections.

The following key is based upon morphological characters only, while the treatment of the genera is in a phyllogenetic order.

THE OHIO JOURNAL OF SCIENCE 55(5): 279, September, 1955.

KEY TO THE TRACHYPHLOEINI.

1. Nasal plate large, smooth, triangular, limited by an abrupt ridge 2
1'. Nasal plate very small or not visible, no limiting ridge present 4
2. Metathoracic episterna visible; suture at least moderately visible ..... 1. *Trachyphloeus* Germar
2'. Metathoracic episterna concealed, no suture apparent; the eyes with a distinct orbital groove 3
3. Scrobes lateral, long, passing directly backward and enclosing the eyes; all tibiae strongly mucronate ..... 2. *Chaetechus* Horn
3'. Scrobes superior, very short and deep, not reaching the eyes; only anterior and middle tibiae mucronate ..... 4. *Cercopeus* Schönherr
4. Scrobes lateral, long, passing backward and below the lower angle of the eyes; all tibiae feebly mucronate ..... 3. *Chaetechidius* n. gen.
4'. Scrobes superior, very short, and deep, not reaching the eyes; only anterior and middle tibiae mucronate at apex 5
5. Rostrum short and broad, prementum not emarginate on apical margin; only the labial palps visible, (fig. 15). 5. *Pseudocercopeus* n. gen.
5'. Rostrum longer, narrow, almost cylindrical, prementum broadly emarginate; both labial and maxillary palps visible, (fig. 12) ..... 6. *Cercopedius* n. gen.

1. **Trachyphloeus** Germar

Trachyphloeus Germar 1817, p. 339. LeConte and Horne 1876, p. 78. Blatchley & Leng 1916, p. 114.

Rostrum as long as the head, stout, subquadrangular, nearly straight, transversely sulcate at base; apex with a prominent triangular nasal plate and limiting ridge. Scrobes lateral, deep, moderately arcuate and passing directly backward to the eye. Antennal scape nearly or attaining the apical margin of the prothorax; funicle 7-segmented, first segment elongate and robust, second segment slender longer than third, segment 3-7 short, 5-7 moniliform; club oval. Eyes small, rounded, lateral, coarsely granulated and with a distinct orbital groove. Prothorax transverse, much wider than long; ocular lobes absent. Scutellum not visible. Elytra oval. Ventral side with the metathoric episterna present, moderate; the suture distinct throughout its entire length. Metasternum short. Intercoxal lobe of first abdominal segment broad, truncate; second segment not as long as 3 and 4 united; first abdominal suture nearly straight. All tibiae usually mucronate (middle tibiae not mucronate in *davisi*); armed with several tooth-like spines at the apices. Claws free and divergent.

Genotype. *Curculio scabriculus* Linneaus by monotypy.

KEY TO THE SPECIES OF *Trachyphloeus*.

1. Anterior tibiae with 3 or 4 distinct, and 2 or more shorter spines at the apex ..... *bifoveolatus* (Beck)
1'. Anterior tibiae with only 2 spines at the apex ..... *davisi* Blatchley

Trachyphloeus bifoveolatus (Beck)

Curculio bifoveolatus Beck 1817, p. 22.
Trachyphloeus bifoveolatus (Beck), Bedel 1883, p. 41. Buchanan 1937, p. 312. Brown 1940, p. 77; 1950, p. 202.
Trachyphloeus asperatus of Horn (nec Boheman) 1876, p. 79.

Oblong-oval, robust; reddish brown to black; densely clothed with a clay-like coating of silver-gray scales; the prothorax with a few scattered recurved clavate scales, the elytra with similar but larger scales in a single row on each interval.

Rostrum robust, slightly shorter than the prothorax, with a deep median groove and a deep transverse depression at the base. Antennal scape nearly reaching anterior margin of the prothorax. Head rounded. Eyes elliptical, very feebly convex.

Prothorax three-fifths wider than long, the sides strongly rounded in the basal two-thirds, then strongly constricted to the apex; pronotum with a longitudinal groove and lateral foveae which are frequently obscured by the crust. Scutellum not visible.

Elytra oblong-oval, about one-fourth longer than broad, the sides rounded to apex; striae not impressed, strial punctures large, round and deep; intervals flat; sculpture of elytra usually completely obscured by the crust.

Ventral side with the sculpture completely obscured by the crust. Anterior tibiae straight; with 3-4 larger and two or more shorter spines at the apex. All tibiae mucronate. Claws free and divergent.

Length 3.4-3.7 mm., width 2.0-2.3 mm.

Type locality Europe.

Distribution. Europe, Canary Islands, Nova Scotia to British Columbia, south to New York and Oregon.

Material examined. Numerous examples from localities in Maine, Nova Scotia, New York, Oregon and British Columbia. In all, 350 examples were studied, all were female. The Maine examples were under rocks in open meadows, while those from Oregon (Portland), were migrating across sidewalks and streets in October.

This species can be separated from *davisi* by the characters mentioned in the key.

Trachyploeus davisi Blatchley

Trachyphloeus davisi Blatchley, Blatchley & Leng 1916, p. 115.

Oblong-oval, robust; black, with the antennae and legs dull reddish brown; densely clothed with dark brown and silver gray scales, the latter in a broad median line and lateral stripes each side of the pronotum and covering the even elytral intervals, forming oblong spots on the third, fifth and seventh intervals, and with scattered clavate erect, feebly recurved scales which are also in a single line on each elytral interval.

Rostrum robust, slightly shorter than the prothorax and with a distinct median groove. Antennal scape attaining anterior margin of the prothorax. Head convex. Eyes elliptical, coarsely granulated and feebly convex.

Prothorax almost twice as wide as long, the sides strongly rounded; pronotum without a median impressed line; the sculpture completely concealed by the crust. Scutellum not visible.

Elytra oblong-oval, the sides feebly divergent to the apical third, then broadly rounded to apex; striae fine, indistinctly punctured; intervals flat.

Ventral side with sculpture completely obscured. Anterior tibiae straight, expanded at apex, and with two spines. Claws free and divergent.

Length 3.5-3.8 mm., width 2.1-2.4 mm.

Type locality. Ft. Wadsworth, Staten Island, New York.

Distribution. Known only from Staten Island and Long Island.

Material examined. Holotype, ♀, Ft. Wadsworth, Staten Island, N. Y., IV–27–11, W. T. Davis in the Blatchley Collection at Purdue University. Other examples without definite data other than a "Long Island" locality label. All examples were ♀.

This species may be separated from *bifoveolatus* (Beck) by the characters mentioned in the key.

2. **Chaetechus** Horn

Chaetechus Horn 1876, p. 77. Blatchley & Leng 1916, p. 113.

Rostrum stout, quadrangular, the sides subparallel, nearly as long as the head; the nasal plate smooth, triangular, limited by a prominent ridge. Scrobes lateral, deep, passing backward and surrounding the eyes. Antennae stout; scape robust, feebly arcuate, attaining apical margin of the prothorax. Funicle 7-segmented, the first two segments elongate, the first very robust; 3–7 moniliform; club oval. Eyes small, with round and a distinct orbital groove. Prothorax transverse. Scutellum very minute. Elytra oblong-oval, the sides subparallel at middle. Ventral side with the second abdominal segment longer than either the third or fourth. First abdominal suture feebly arcuate. Inter-coxal lobe of first abdominal segment very broad and short. All tibiae mucronate at apex. Claws free and divergent.

Genotype. Chaetechus setiger Horn which is a synonym of *Trachyphloeus asperatus* Boheman.

Chaetechus asperatus (Boheman)

Trachyploeus asperatus Boheman 1844, p. 116.
Chaetechus setiger Horn 1876, p. 78. Blatchley & Leng 1916, p 114. Blackwelder 1948, p. 46.
Chaetechus asperatus (Boheman), Blackwelder 1948, p. 46.

Oval, convex; reddish brown; covered with a crust of gray scales and a clay-like substance, and suberect, slightly recurved, clavate scales, the latter in a single row on each elytral interval.

Rostrum short and broad, little more than half as long as prothorax, the sides subparallel; densely punctured and with an indistinct median line, the sculpture frequently concealed by the clay-like coating. Eyes round and strongly convex.

Prothorax one-fourth wider than long, the sides divergent to apical third, then constricted to the apex; pronotum moderately convex, densely punctured, a slight post-apical and short transverse impression slightly behind the middle; sculpture obscured by the crust; surface with the clavate scales rather sparse. Scutellum very minute.

Elytra oval, three-fourths as wide as long, the sides parallel behind humeri apical third, then convergent to the apex; striae feebly impressed, strial punctures round, very deep; intervals flat, very minutely punctured; sculpture of surface concealed by the crust.

Ventral side with a few erect setae and coarsely, but not densely punctured. The legs clothed with clavate scales. Femora unarmed. Anterior tibiae slightly curved. All tibiae mucronate at apex.

Length 2.3–2.6 mm. width 1.2–1.4 mm.

Type locality. Of *asperatus* Boheman "America borealis ad Boston", of *setiger* Horn "Massachusetts".

Distribution. Maine, New Hampshire, and Massachusetts.

Material examined. Maine: Harrington, VI–II–34, in the author's collection and the United States National Museum. Examples also were studied from New Hampshire and Massachusetts without precise locality. Thirty-five examples were studied, all were female.

3. **Chaetechidius** n. gen.

Rostrum stout, quadrangular, the sides subparallel, shorter than the head, the nasal plate and limiting ridge absent. Scrobes lateral, long, passing backwards and beneath the lower angle of the eyes. Antennal scape slender, arcuate and attaining the apical margin of the prothorax. Funicle 7-segmented, the first sement very elongate and robust, the second elongate and slender, 3–7 moniliform; club ovate. Eyes small, round, and surrounded by a deep orbital groove. Prothorax transverse, much wider than long; ocular lobes absent. Scutellum not visible. Elytra oval, the sides rounded. Metaepisterna concealed. Intercoxal lobe on the first abdominal segment broad and truncated. Second abdominal segment longer than the third and fourth combined. First abdominal suture straight. Femora unarmed. All tibiae feebly mucronate. Claws free and divergent.

Genotype. Chaetechidius speciosus n. sp.

The genotype resembles a *Chaetechus* but may be readily separated by the absence of a nasal plate and limiting ridge, and the more rounded elytra.

Chaetechidius speciosus n. sp.

Female. Oval, convex: black, with the antennae, tarsi and 3–5 abdominal segments reddish brown; densely clothed with very delicate, overlapping, white radiate-pectinate scales and scattered erect, rod-like setae which on the elytra are in a single line on each interval.

Rostrum short, broad, almost three-fifths as long as the prothorax; very densely rugosely punctured and with a shallow median groove from the base to antennal insertion; head and rostrum continuous, not separated by a transverse groove. First segment of funcile elongate and very robust, the second segment elongate and about one-half as long as first, 3–7 moniliform. Club oval, densely pubescent. Eyes small, round and very strongly convex.

Prothorax more than one-half wider than long, the sides rounded from base to apex; pronotum convex, densely, coarsely, deeply punctured. Scutellum not visible.

Elytra oval, one-fifth wider than long, the sides rounded from base to apex; striae unimpressed, strial punctures round, deep and closeset; intervals flat sparsely, finely punctured.

Ventral side very sparsely clothed with semierect narrow scales. The thoracic sterna and the first abdominal segment rugosely, rather coarsely punctured, the remaining segments very sparsely and finely punctured. Anterior tibiae straight, the apex with numerous long, slender, sharp spines. All tibiae feebly mucronate.

Length 2.4 mm., width 1.5 mm.

Male. Unknown.

Type locality. Colorado Springs, Colorado.

Distribution. Known only from Colorado Springs, and Greely, Colorado.

Type material. Holotype, ♀, Colorado Springs, Colorado, X-10, in the author's collection. Other examples have been seen from Colorado Springs and Greely, Colorado. All were female. The holotype was found under a rock in a grassy area.

This species resembles, superficially, *Chaetechus asperatus* (Boheman), but differs in being more oval, with the sides of the elytra more rounded, in the absence of the nasal plate and limiting ridge and the erect, rod-like, instead of clavate, setae.

4. **Cercopeus** Schönherr

Cercopeus Schönherr 1843, p. 154. Lacordaire 1863, p. 192. Seidlitz 1865, p. 277. Pierce 1913, p. 422. Blatchley & Leng 1916 p. 109.

Cercopius Gemminger & Harold 1871, p. 2287. (emend.)

Cerpoceus Lacordaire 1863, p. 192. (err.)

Rostrum slightly longer and narrower than the head, feebly curved; nasal plate smooth, triangular, limited by a prominent ridge. Scrobes superior, short, deep, not attaining the eyes. Funicle 7-segmented; the first two segments elongate, the first very robust, the outer segments shorter or moniliform. Eyes small, rounded, with a distinct orbital groove. Ocular lobes absent. Elytra oval, humeral angles rounded, the sides broadly rounded. Metaepisternum concealed, the metasternum short. Intercoxal lobe of first abdominal segment large, broad and truncated. Anterior and middle tibiae feebly mucronate. Hind tibiae usually with two fixed spurs and the corbels open. Tarsal claws free and divergent.

Genotype. Peritelus chrysorrhoeus Say, by original designation.

KEY TO THE SPECIES OF *Cercopeus*.

1. Prothorax strongly constricted apically (fig. 11); pronotal punctures small, moderately dense, but not confluent; hind tibiae with the spurs very short in the ♂, one of them wanting in the ♀; Texas ..... *isquilus* n. sp.
1'. Prothorax slightly or but very feebly constricted (fig. 16); pronotal punctures more elongate and at least slightly confluent; hind tibiae with the spurs moderately long 2
2. Pronotum with a median longitudinal ridge apically and at middle of base; eyes very convex and prominent; Mich., Ill. ..... *schwarzi*, n. sp.
2'. Pronotum without such modification 3
3. Outer apical angle of anterior tibiae sharply produced; spines in distal comb of hind tibiae short; antennae very robust; middle of pronotum with a distinct tumidity; a robust, dark species; Mass., Pa., Va., N. C. ..... *maspavancus* n. sp.
3'. Outer apical angle of anterior tibiae not produced; antennae more slender; variable species 4
4. Disc of pronotum strigose, the individual punctures nearly effaced 5
4'. Disc of the pronotum with the punctures present (though they may be strongly elongated and in part confluent; middle of pronotum not tumid 6
5. Larger 4.5–5.5 mm.; pronotal striae rather fine, not widely separated; outer apical angle with a small spine above apex; darker and with no distinct color pattern; S. C., Ga., Ala. ..... *strigicollis* n. sp.
5'. Smaller, 3.0–3.6 mm.; pronotal striae very coarse, widely separated, outer apical angle without a small spine above apex; with a distinct color pattern; Ark. ..... *clispus* n. sp.
6. Dorsal edge of rostrum, in profile, concave at middle; a tumidity between antennal insertion and on vertex of head above eyes (fig. 14); apex of rostrum swollen; Va. *simius* n. sp.
6'. Not as above 7
7. Anterior tibiae with a prominent spine on the outer edge some distance above the apex; eyes elliptical, moderately convex, rostrum thinner; Ohio, N. Y., Pa., N. J., Md., D. C., Va., S. C., Ala., Tex. ..... *chrysorrhoeus* (Say)
7'. Anterior tibiae without such a prominent spine, the spines evenly distributed; eyes rounded, strongly convex; rostrum thicker 8
8. Smaller, 2.0–3.0 mm.; prothorax very finely, closely, deeply punctured, the intervals between the punctures raised giving a granular appearance; 8th sternite of ♀ narrowed at apex, truncated (fig. 18); Ill., Ia., Mo., Ark. ..... *chisaius* n. sp.
8'. Larger, 3.0–3.5 mm.; prothorax very coarsely, closely punctured, the surface not granular in appearance; 8th sternite of ♀ broadly rounded at apex, not narrowed (fig. 19); Ohio ..... *confusor* n. sp.

Cercopeus isquitus n. sp.

Figures 1 and 11

Female. Oval, strongly convex; reddish brown with the antennae and tarsi slightly paler; densely clothed with pale brown, dark brown and white scales, the white scales condensed laterally on the prothorax, in spots on the humeri, at the base of the 6th interval and in an indefinite oblique white band at declivity; head, prothorax and elytra with erect, slightly recurved setae, those on the elytra in a single row on each interval.

Rostrum short, broad, three-fifths as long as prothorax; nasal plate very prominent, very oblique in relation to the remainder to the dorsal edge; dorsal edge, in profile, feebly convex above the eyes. Head strigose, the punctures confluent. Eyes moderately convex, round and prominent.

Prothorax rectangular, one-fifth wider than long, the sides subparallel at middle, strongly rounded to base and constricted to apex; pronotal punctures small, moderately dense, never confluent. Scutellum very small, nearly invisible, densely clothed with white scales.

Elytra oval, nearly as wide as long, the sides strongly rounded; striae moderately impressed, strial punctures feeble; intervals flat.

Ventral side with the thoracic sternites densely clothed with white scales; the abdominal segments convex; sparsely clothed with fine suberect setae and very sparsely punctured, first abdominal suture straight; the first abdominal segment with a depression at middle of apical margin. Hind tibiae of female with only one fixed spur. Anterior tibiae straight, slightly expanded at outer apical angle.

Length 2.7 mm., width 1.5 mm.

Male. Differs from the female in the presence of two spurs on the hind tibiae instead of one, and abdominal segments 1 and 2 broadly depressed at middle. Length 2.7 mm., width 1.5 mm.

Type locality. Washington Co., Texas.

Distribution. Washington and Walker Counties, Texas.

Type material. Holotype, ♀, Washington Co., Texas in the author's collection, allotype, ♂, and one paratype, ♀, Hempstead, Texas in the United States National Museum.

This species is not closely related to any of the other species. It may be readily separated by the rectangular prothorax, the non-confluent punctures and the shorter, blunt spurs on the hind tibiae. The holotype was found in woodland litter.

Cercopeus schwarzi n. sp.

Figure 2

Female. Elongate, oval, convex; dark reddish brown densely clothed with brown scales and erect, feebly recurved setae, those on the elytra arranged in single rows on each interval, without evident color pattern.

Rostrum short, broad slightly more than one-half as long as prothorax; feebly sulcate at middle, the sulcus nearly obscured by scales; dorsal edge, in profile, convex. Head strigose; with a shallow transverse depression above the eyes. Eyes moderately convex, oval, prominent.

Prothorax slightly wider than long, sides rounded from base to apex; pronotum with a prominent keel in apical third and at middle of base; coarsely, deeply, closely punctured. Scutellum very small, nearly invisible.

Elytra elongate-oval, one-fourth longer than wide, the sides arcuate from base to apex; striae impressed, the strial punctures very feeble; intervals flat or just preceptibly convex.

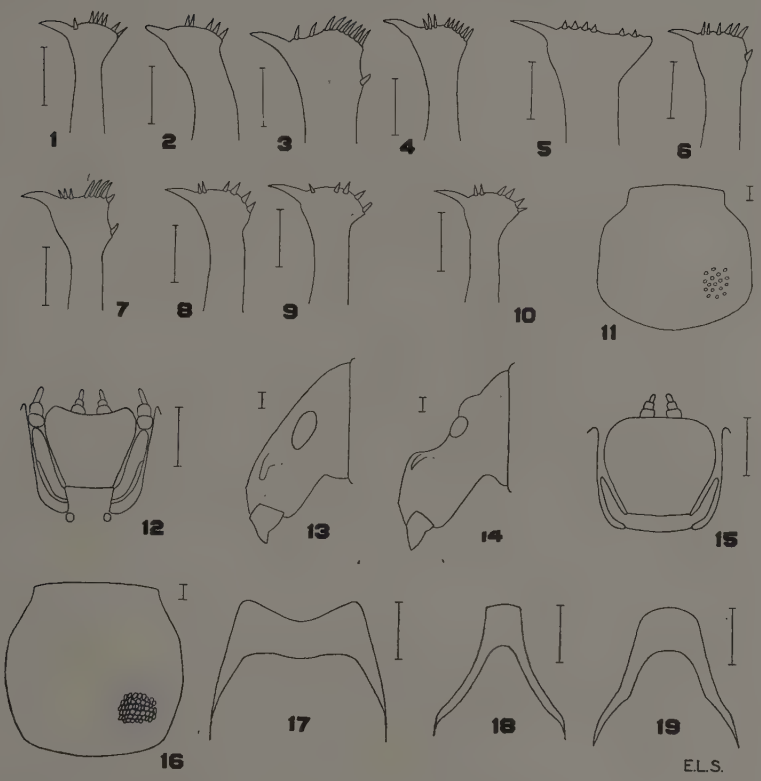
Ventral side with the thoracic sternites densely clothed with scales, the abdominal segments sparsely clothed with brown scales and recumbent setae and a few sparse punctures. First abdominal suture strongly curved. Hind tibiae of ♀ with two fixed spurs. Anterior tibiae strongly curved, the outer apical angle not produced.

Length 3.6-mm., width 1.75 mm.

Male. Unknown.

Type locality. Detroit, Michigan.

Distribution. Michigan and Illinois.



1. Apex of right anterior tibiae of *Cercopeus isquitus* n. sp., ♀.
2. Same of *Cercopeus schwarzi* n. sp., ♀.
3. Same of *Cercopeus strigicollis* n. sp., ♀.
4. Same of *Cercopeus clispus* n. sp., ♀.
5. Same of *Cercopeus maspavancus* n. sp., ♀.
6. Same of *Cercopeus simius* n. sp., ♀.
7. Same of *Cercopeus chrysorrhoeus* (Say), ♀.
8. Same of *Cercopeus chisaius* n. sp., ♀.
9. Same of *Cercopeus confusor* n. sp., ♀.
10. Same of *Cercopedius artemisiae* (Pierce), ♀.
11. Outline of prothorax of *Cercopeus isquitus* n. sp.
12. Prementum of *Cercopedius artemisiae* (Pierce).
13. Profile of rostrum of *Cercopeus chrysorrhoeus* (Say).
14. Same of *Cercopeus simius* n. sp.
15. Prementum of *Pseudocercopeus setosus* n. sp.
16. Outline of prothorax of *Cercopeus chrysorrhoeus* (Say).
17. Eighth sternite of ♀ of *Cercopeus chrysorrhoeus* (Say).
18. Same of *Cercopeus chisaius* n. sp.
19. Same of *Cercopeus confusor* n. sp.

Line represents 1/6 mm.

Type material. Holotype, ♀, Detroit, Michigan, Hubbard & Schwarz, in the United States National Museum, 3 paratypes: 2 ♀, Detroit, Mich., in the United States National Museum; 1 ♀, same data as the holotype, in the author's collection. Length of paratypes 3.2–3.4 mm., width 1.6–1.8 mm. Examples have also been seen from Chicago, Illinois, but are not included as a part of the type series.

This species differs from *C. chrysorrhoeus* (Say) in the prominent pronotal carinae and absence of any color pattern. From the other species it may be separated by the characters mentioned in the key.

Cercopeus maspavancus n. sp.

Figure 5

Female. Oval, convex; dark reddish brown; densely clothed with light and dark brown scales and recumbent setae; a discal spot of dark scales on the pronotum and on the elytra, the elytra with an oblique band of pale scales from humeri to declivity; the setae arranged in a single row on each elytral interval.

Rostrum short, broad, only one-half as long as the prothorax; dorsal edge, in profile, feebly convex; the nasal plate very prominent and oblique. Antennae very robust. Head closely and coarsely punctured. Eyes convex, oval, not prominent.

Prothorax wider than long, the sides evenly rounded from base to apex; pronotum coarsely, deeply, densely punctured middle of pronotum distinctly tumid. Scutellum not visible.

Elytra oval; sides rounded from base to apical third, then strongly convergent to apex; striae strongly impressed, strial punctures very small; intervals very feebly convex.

Ventral side sparsely clothed with scales and fine recumbent setae. Abdominal segments moderately densely punctured. First abdominal suture strongly curved at middle. Hind tibiae of ♀, with two fixed spurs; spines of distal comb of hind tibiae short. Anterior tibiae straight, the outer apical angle sharply produced.

Length 4.5 mm., width 2.5 mm.

Type locality. Shrewsbury, Worcester Co., Massachusetts.

Distribution. Massachusetts, Pennsylvania, Virginia, and North Carolina.

Type material. Holotype, ♀, Shrewsbury, Mass., in the author's collection, 4 paratypes: 1 ♀, Chincoteagua, Va., X-8-34, H.S. Barber, in the author's collection; 1 ♀, Delaware Co., Pa., X-19-13, Kaeber, 1 ♀, Southern Pines, N.C., IV-2-11, A. H. Manee, 1 ♀, Southern Pines, N.C., IV-20-11, A. H. Manee in the United States National Museum. Length of paratypes 4.0–4.5 mm., width 2.1–2.3 mm.

This species can be easily separated from all other species by the sharply produced outer apical angle of the anterior tibiae and the short spines in the distal comb of the posterior tibiae. The holotype was taken by sifting woodland litter.

Cercopeus strigicollis n. sp.

Figure 3

Female. Elongate-oval, convex; dark reddish brown; densely clothed with dark brown, and gray scales and erect recurved setae; the gray scales condensed on the sides of the elytra and prothorax; the erect setae in a single row on each elytral interval; no definite color pattern evident.

Rostrum short, broad, slightly more than one-half as long as the prothorax; dorsal edge, in profile, convex, nasal plate not obliquely set off. Head prominently strigose. Eyes convex, oval, not prominent.

Prothorax one-fifth wider than long, the sides evenly rounded from base to apex; pronotum longitudinally strigose, the punctures nearly effaced; middle of pronotum distinctly tumid. Scutellum very small but visible.

Elytra elongate-oval, one-third longer than wide; the sides rounded from base to apex; striae strongly impressed, strial punctures rectangular; intervals flat.

Ventral side sparsely clothed with gray scales and fine suberect setae. Abdominal segments smooth, very sparsely irregularly punctured. First abdominal suture very feebly arched. Hind

tibiae of ♀ with two fixed spurs. Anterior tibiae straight; the outer apical angle not produced but with a spine some distance above the apex.

Length 4.25 mm., width 2.25 mm.

Male. Unknown.

Type locality. Marion, Marion County, South Carolina.

Distribution. South Carolina, Georgia and Alabama.

Type material. Holotype, ♀, Marion, S.C., II–12–35, in the author's collection; 5 paratypes: 2 ♀, Experiment, Ga., II–25–36, T. Bissell, in the author's collection; 2 ♀, Florence, S.C., II–13–39, 1 ♀, Clemson College, S.C., III–26–33, J. A. Berley, in the United States National Museum. Several examples have been seen from Alabama, but are not included as a part of the type series. Length of paratypes 4.5–4.7 mm., width 2.4–2.6 mm. The holotype was found in woodland litter. The paratypes from Florence, S.C. were in woods trash. The examples from Experiment, Ga. were in dead leaves.

This species is nearest *C. clispus* no. sp. from which it differs in larger size, presence of the spine some distance from the outer apical angle of the anterior tibiae, the indistinct color pattern and the more finely strigose pronotum. It differs from *C. chrysorrhoeus* (Say) in the strigose, slightly tumid pronotum and indistinct color pattern.

Cercopeus clispus n. sp.

Figure 4

Female. Elongate-oval, convex; dark reddish brown; densely clothed with gray and dark brown scales and erect setae; the gray scales scattered on pronotum and in an oblique band on the elytra from the humeri to the declivity; the erect setae on the elytral intervals in a single row.

Rostrum short, broad, about one-half as long as prothorax, dorsal edge, in profile, arcuate from base to apex. Head strigose. Eyes oval, not at all prominent.

Prothorax as long as wide, sides feebly rounded, almost parallel except at base and apex; pronotum very coarsely, deeply strigose, the strigae not longitudinal; pronotum without a tumidity. Scutellum not visible.

Elytra oval, more than one-third longer than wide, the sides rounded from base to apex; striae very feebly impressed, strial punctures deep, rounded, and very small; intervals flat.

Ventral side with the thoracic sterna densely clothed with broad gray scales, sparsely clothed with fine recumbent setae. Abdominal segments smooth, convex, very sparsely and coarsely punctured. First abdominal suture feebly arcuate at the middle. Hind tibiae of ♀ with two fixed spurs at apex. Anterior tibiae straight; not sharply expanded at the outer apical angle, without a distinct spine on the outer edge some distance from the apex.

Length 3.3 mm., width 1.6 mm.

Male. Unknown.

Type locality. Ozark, Franklin County, Arkansas.

Distribution. Arkansas.

Type material. Holotype, ♀, Ozark, Ark., II–20–50, in the author's collection; 5 paratypes: 2 ♀, Johnson Co., Ark., II–26–48, in the author's collection; 3 ♀, Johnson Co., Ark., in the Entomological Collection, University of Arkansas, Fayetteville. Length of paratypes 3.3–3.5 mm., width 1.6–1.7 mm.

This species is related to *C. strigicollis* n. sp. but may be easily separated from it by the much coarser pronotal striae, the absence of the spine on the outer edge at some distance from the outer apical angle, and smaller size. The type was found in woodland litter. Those from Johnson, Co., Ark. were all from orchard trash.

Cercopeus simius n. sp.

Figures 6 and 14

Female. Oval, convex; reddish brown; densely clothed with pale brown scales and erect, slightly recurved setae, the latter in a single row on each elytral interval.

Rostrum short, moderately broad, three-fifths as long as prothorax; dorsal edge, in profile, deeply concave between eyes and insertion of the antennae; a prominent tumidity between the

antennal insertion; apex of rostrum swollen, wider than middle. Head strigose, with a prominent tumidity on vertex. Eyes round, convex and placed more dorsally than usual.

Prothorax scarcely wider than long, the sides evenly rounded from base to apex; pronotum coarsely, closely, deeply punctured. Scutellum not visible.

Elytra oval, sides rounded from base to apex, nearly one-third longer than wide; striae deeply impressed, strial punctures small rounded and deep; intervals flat.

Ventral side very sparsely clothed with scales and fine recurved setae. Abdominal segments very smooth, very sparsely punctured; first abdominal segment with a concave depression at middle of apical margin. First abdominal suture strongly arcuate. Hind tibiae of ♀ with two fixed spurs. Anterior tibiae straight not expanded at outer apical angle, with a prominent spine some distance from apex.

Length 3.2 mm., width 1.7 mm.

Male. Unknown.

Type locality. Stone Creek, Lee Co., Virginia.

Distribution. Lee Co., Va.

Type material. Holotype, ♀, Stone Creek, Lee Co., Va., VII–31, Hubbard & Schwarz, in the United States National Museum.

Easily separated from all the other species by the peculiar characters of the rostrum, which are very prominent. The placement of the eyes is also rather peculiar.

Cercopeus chrysorrhoeus (Say)

Figures 7, 13, 16 and 17

Peritelus chrysorrhoeus Say 1831, p. 13. Boheman in Schöherr 1834, p. 517. LeConte 1857, I, p. 274.

Cercopeus chryorhoeus Schönherr 1843, I, p. 155. (err.)

Cercopeus chrysorhoeus Blatchley & Leng 1916, p. 113. (err.)

Female. Oval, convex; reddish brown; densely covered with pale, and dark brown scales and inclined setae; the pale brown scales condensed in a line on the sides of the pronotum and in an oblique line on the elytra from the base to the declivity; the setae on the elytra arranged in a single row on each elytral interval.

Rostrum short, broad, two-thirds as long as prothorax; dorsal edge, in profile, evenly arcuate from eyes to apex. Head strigose, a feeble transverse depression above the eyes. Eyes moderately convex, elliptical, not very prominent.

Prothorax about as long as wide, the sides rounded from base to apex; pronotum coarsely, closely punctured; middle of pronotum not tumid. Scutellum not visible.

Elytra oval, almost one-third longer than wide; the sides rounded from base to apex; striae moderately impressed, the strial punctures very small, deep, often obscured by scales; intervals flat.

Ventral side sparsely clothed with gray scales and sub-erect setae. The scales nearly absent from the abdominal segments which are smooth and shining, feebly convex, sparsely punctured. First abdominal suture arcuate at middle. Hind tibiae of ♀, with two fixed spurs at the apex. Anterior tibiae straight, the outer edge with a spine some distance from the outer apical angle, the latter not produced outward.

Length 3.6 mm., width 1.75 mm.

Male. Unknown.

Type locality. Pennsylvania. Type destroyed.

Neotype locality. Rhododendron Hollow, Clear Creek, Goodhope, Twp., Hocking Co., Ohio.

Distribution. Massachusetts to Ohio, south to South Carolina, Alabama and Texas.

Neotype material. Neoholotype, ♀, Clear Creek, Hocking Co., Ohio, III-24-53, E. L. Sleeper, in the author's collection, 3 neoparatypes: 1 ♀, Clear Creek, Hocking Co., II–21–53, E. L. Sleeper. 4 ♀, same locality, IV–2–55, E. L. S., 1 ♀, Pittsfield, Mass., 1 ♀, Buckhannon, W. Va., all in the author's collection.

This species can be separated from the other species by the non-strigose, non-tumid pronotum and the presence of the spine on the outer edge of the anterior tibiae above the outer apical angle.

All examples were taken by sifting in predominantly Oak woods.

Cercopeus chisaius n. sp.

Figures 8 and 18

Female. Oval, convex; reddish brown; densely clothed with gray and dark brown scales and short erect setae, the elytra with an oblique band of dark brown scales from the base to the declivity; the setae in a single row on each interval.

Rostrum short, broad, more than one-half as long as prothorax, dorsal edge, in profile, arcuate from eyes to nasal plate, the latter very oblique. Head coarsely, closely punctured. Eyes very strongly convex, rounded, very prominent.

Prothorax a little wider than long, the side arcuate from base to apex; pronotum very finely, closely, deeply punctured, the intervals between the punctures raised, giving the surface a granular appearance, not tumid at middle. Scutellum not visible.

Elytra oval, one-fifth longer than wide, the sides strongly rounded from base to apex; striae feebly impressed, strial punctures very small, deep and round; intervals flat.

Ventral side with the thoracic sterna densely clothed with opaque scales. Abdominal segments feebly convex, shining, very sparsely, finely punctured, very sparsely clothed with fine recumbent setae. Hind tibiae of ♀, with two fixed spines at apex. The anterior tibiae straight, not expanded at outer apical angle, without trace of a spine some distance above the apex on the outer edge. Eighth sternite of ♀ narrowed at apex, truncated (fig. 18).

Length 2.6 mm., width 1.4 mm.

Male. Unknown.

Type locality. West Vienna, Illinois.

Distribution. Illinois, Iowa, Missouri and Arkansas.

Type material. Holotype, ♀, West Vienna, Ill., XII–30–32, in the author's collection; 8 paratypes: 1 ♀, West Vienna, Ill., III–30–32, 2 ♀, Homer, Ill., III–30–07, Hart & Hood, in the author's collection; 4 ♀, Homer, Ill., III–30–07, Hart & Hood, in the United States National Museum; 1 ♀, "Ark." in the Entomological Collection of the Illinois State Natural History Survey. Other examples have been seen from Illinois, Iowa, Missouri and Arkansas, but are not a part of the type series. Length of paratypes 2.3–2.8 mm., width 1.3–1.7 mm.

This species is related to *C. confusor* n. sp. as is shown by the characters of the 8th sternite of the ♀ and the anterior tibiae, but may be separated from it by the smaller size, and finer denser pronotal punctures. From *chrysorrhoeus* it differs in the smaller size, finer pronotal punctures, narrower, non-emarginate 8th sternite of ♀ and the absence of the spine on the outer edge of the anterior tibiae above the apical angle. The holotype was taken by sifting in soil cover near the edge of a woods.

Cercopeus confusor n. sp.

Figures 9 and 19

Female. Oval, convex; reddish brown; densely clothed with gray, medium brown and dark brown scales, and erect slightly recurved setae; the gray and medium brown scales in a discal patch on the elytra, the erect setae in a single row on each interval.

Rostrum short, broad, slightly more than one-half the length of the prothorax; dorsal edge, in profile, strongly arcuate from eye to nasal plate which is strongly oblique. Head coarsely, deeply, closely punctured. Eyes elliptical, strongly convex and prominent.

Prothorax nearly as long as broad; the sides rounded from base to apex: pronotum closely, coarsely and deeply punctured; not tumid at middle. Scutellum not visible.

Elytra oval, one-fourth longer than wide; the sides rounded from base to apex; striae feebly impressed, strial punctures small, round and deep; intervals flat.

Ventral side with the thoracic sterna moderately clothed with gray scales and scattered recumbent setae. Abdominal segments smooth feebly convex, very sparsely, finely punctured, sparsely clothed with fine recumbent setae. First abdominal suture moderately arched at middle. Hind tibiae of ♀ with two fixed spurs at apex. Anterior tibiae straight, not expanded at the outer apical angle and without a single spine some distance from the apex. Apex of 8th sterna of ♀ broadly rounded (fig. 19).

Length 3.2 mm., width 1.7 mm.

Male. Unknown.

Type locality. Roosevelt Lake, Nile Twp., Scioto Co., Ohio.

Distribution. Known only from the type locality.

Type material. Holotype, ♀, Roosevelt Lake, Scioto Co., Ohio, XII–30–53, E. L. Sleeper, in the author's collection; 2 paratypes: 1 ♀, same data as holotype, in the author's collection; 1 ♀, same locality and date as holotype, R. E. Woodruff, in the R. E. Woodruff Collection. All examples were taken by sifting woodland litter in a high, dry oak-hickory woods.

This species is near *C. chisaius* n. sp. and maybe separated by the characters mentioned in the discussion following the description of that species.

Pseudocercopeus n. gen.

Rostrum longer and slightly narrower than the head, straight; apex of the rostrum without indication of a nasal plate or limiting ridge. Prementum not emarginate on apical margin, labial palpi only visible. Scrobes superior, short, deep and convergent above. Antennal scape attaining the apical margin of the prothorax. Funicle 7-segmented, the first two elongate, the first segment more robust, the third segment longer than broad, 4–7 moniliform. Club elongate-oval. Eyes rounded, convex, not surrounded by an orbital groove. Prothorax wider than long, the base and apex truncate; ocular lobes absent. Scutellum not visible. Elytra elongate; humeri effaced; sides arcuate. Ventral side with the metaepisterna concealed, the intercoxal lobe of the first abdominal segment broad and truncated. Second segment short, a little longer than the third, the third and fourth subequal. First abdominal suture feebly arcuate. Femora unarmed. Anterior and middle tibiae feebly mucronate. Claws free and divergent.

Genotype. Pseudocercopeus setosus n. sp.

The genotype resembles an elongate *Cercopeus* but lacks the orbital groove, the nasal plate and the limiting rige. It may be easily separated from *Cercopedius* by the characters mentioned in the key.

Pseudocercopeus setosus n. sp.

Figure 15

Male. Elongate, convex; reddish brown; densely clothed with white, light, and dark brown round and slightly overlapping scales and semierect setae which on the elytra are in a single row on each elytral interval.

Rostrum short, broad, about three-fifths as long as prothorax, the sides feebly convergent from the base to the apex; dorsal edge, in profile, continuous with the head, arcuate from the apical margin of the prothorax to the apex; finely, sparsely punctured, the median line without a groove. Antennal scape feebly clavate, slender; first segment of funicle elongate, robust about one-third longer than the second, the second one-half longer than the third. Club sparsely pubescent. Head very finely and sparsely punctured. Eyes round, moderately convex, and very coarsely granulate.

Prothorax about one-fifth wider than long, the sides rounded to a feeble apical constriction; pronotum convex, smooth, very sparsely and finely punctured, the scales obscuring the punctures. Scutellum absent.

Elytra elongate, nearly two-fifths longer than wide, the sides feebly arcuate behind middle to apical third, then strongly rounded to the apex; striae not impressed, strial punctures very faint; intervals flat, confusedly, minutely punctured.

Ventral side sparsely clothed with white scales and semierect setae. Abdominal segments convex, very finely, sparsely punctured; the first and second segments feebly impressed at middle. Legs densely clothed with scales and setae. Anterior tibiae straight, with numerous long, sharp spines at the apex.

Length 3.2 mm., width 1.5 mm.

Female. It differs from the male in having the first and second abdominal segments convex and not impressed. Length 3.0 mm., width 1.6 mm.

Type locality. Tempre, Arizona.

Distribution. To present, known only from the type locality.

Type material. Holotype, ♂, and allotype, ♀, Tempre, Ariz., I–31, in the author's collection.

This species resembles an elongate *Cercopeus* but may be readily separated from it by the absence of the nasal plate and limiting ridge and the absence of the orbital groove around the eyes. It may be separated from *Cercopedius* by the shorter rostrum.

Cercopedius n. gen.

Rostrum rather slender, subcylindrical, the sides feebly convergent from base to apex; the apex without a nasal plate and limiting ridge. Prementum broadly emarginate on apical margin, both maxillary and labial palpi visible. Scrobes superior, short, deep and convergent above. Antennal scape slightly arcuate, attaining the apical margin of the prothorax. Funicle 7-segmented, the first two segments elongate subequal, the first segment robust, the second slender, segments 3–4 longer than broad, 5–7 moniliform. Club elongate, acuminate at tip. Eyes round, not surrounded by an orbital groove. Prothorax transverse, a little wider than long; ocular lobes absent. Scutellum not visible. Elytra oblong-oval, the sides broadly rounded from base to apex. Ventral side with the metaepisterna concealed, the intercoxal lobe of the first abdominal segment broad and truncated. Second abdominal segment longer than third and fourth united. First abdominal suture straight. Femora unarmed. Anterior and middle tibiae mucronate. Anterior tibiae straight, the outer apical angle feebly expanded. The apex of each tibiae with several spines. Claws free and divergent.

Genotype. Cercopeus artemisiae Pierce.

This genus can be separated from *Cercopeus* by the longer rostrum and the absence of a nasal plate and limiting ridge. From the genera lacking the nasal plate it may be separated by the characters in the key.

Cercopedius artemisiae (Pierce) n. comb.

Figure 12

Cercopeus artemisiae Pierce 1910, p. 365. Blackwelder 1948, p. 46.
Lepidopus parvulus Van Dyke 1936, p. 78.
Cercopeus parvulus (Van Dyke), Blackwelder 1948, p. 46.

Elongate-oval, convex; black, with the antennae and tarsi reddish brown; densely clothed with round, feebly overlapping white and yellowish brown, finely striate, appressed scales and pale, fine, suberect setae which are widely separated and in a single row on each elytral interval.

Rostrum rather slender, subcylindrical, the sides feebly convergent from base to apex, two-thirds as long as prothorax; finely, sparsely punctured, and with a narrow median groove from base to insertion of antennae. Antennae slender, the funicle with the first segment elongate and robust, one-third longer than the second which is more slender and about one-third longer than the third, segments 3–4 elongate, longer than broad, 5–7 moniliform. Club ovate-acuminate. Head convex and with or without an interocular fovea. Eyes round, feebly convex and coarsely granulate.

Prothorax about one-fifth wider than long, the base and apex feebly constricted, the sides strongly rounded between the constrictions; pronotum convex, very finely sparsely punctured, the punctures obscured by the scales. Scutellum not visible.

Elytra elongate-oval, about two-fifths longer than broad, the sides broadly arcuate from base to apex; striae unimpressed and narrowly denuded giving an appearance of being strongly impressed, strial punctures very fine, shallow and round; intervals flat, finely punctured.

Ventral side rather densely clothed with rounded scales and suberect setae. Abdominal segments convex, finely, sparsely punctured and strongly alutaceous. First abdominal suture straight. Legs densely scaley. Anterior tibiae straight, slightly expanded at outer apical angle, apices with moderately long spines.

Length 2.7–3.5 mm., width 1.4–1.8 mm.

Type locality. Type of *artemisiae* Pierce was from Corvallis, Montana, that of *parvulus* Van Dyke was from Parma, Idaho.

Distribution. Montana, Idaho and Washington.

Material examined. *Montana*: Corvallis, V-09, R. A. Cooley, (holotype and 4 paratypes), in the United States National Museum. *Idaho*: Parma, IV-1-31, H. P. Lanchester, (holotype of *parvulus*), in the Entomological Collection, California Academy of Sciences. *Washington*: 5 ♀, 5 mi. W. College Pl., IV-17-49, G. H. Nelson, in the author's collection and G. H. Nelson Collection; 40 ♀, Pullman, IV-15-40, in the United States National Museum and the author's collection; 20 ♀, Ephrata, V-5-49, F. F. Ritz, in the United States National Museum and the author's collection. The examples from near College Pl., Wash. were beaten from sage brush. The type and paratypes were found feeding on cherry.

LITERATURE CITED

Beck, L. von. 1817. Beitrage zur baierischen Insecten Fauna. Augsburg, Wolff. 45 p. Illus.
Bedel, L. 1883. Fauna d. Coleopteres du bassin de la Seine. II Ann. Soc. Ent. Fr. (6) 3: 33–44.
Blackwelder, R. E. and **R. M. Blackwelder.** 1948. Fifth supplement to the Leng Catalogue of the Coleoptera of America, North of Mexico. 87p. John D. Sherman. Mount Vernon, N. Y.
Blatchley, W. S. and **C. W. Leng.** 1916. Rhynchophora or weevils of Northeastern America. 682 p. The Nature Publishing Company, Indianapolis.
Boheman, C. H. Descriptions in Schönherr, C. J.
Brown, W. J. 1940. Notes on the American distribution of some species of Coleoptera common to the European and North American Continents. Can. Ent. 72: 65–78.
———. 1950. The extralimital distribution of some species of Coleoptera. Can. Ent. 82: 197–205.
Buchanan, L. L. 1937. Notes on Curculionidae (Coleoptera). Journ. Wash. Ac. Sci. 27: 312–316.
Gemminger, M. and **E. von Harold.** 1871. Catalogus Coleopterorum VIII, Curculionidae. p. 2181–2668. Munich.
Germar, E. F. 1817. Magazine der Entomologie, Halle, II, 346 p. Illus.
Lacordaire, P. and **Chapuis.** 1863. Histoire Naturelle des Insectes. Genera des Coleopteres, VI, Curculionides. Paris.
LeConte, J. L. 1859. Editor. American Entomology by Thomas Say. New York. vol. I, 412 p. Illus.
———, and **G. H. Horn.** 1876. The Rhynchophora of America north of Mexico. Proc. Am. Phil. Soc. 15: 1–455.
Leng, C. W. 1920. Catalogue of the Coleoptera of America, north of Mexico. 470 p. John D. Sherman. Mount Vernon, N. Y.
Lona C. 1937. Otiorhynchinae II, Curculionidae. Coleopterorum Catalogus. par. 160. p. 229–412.
Pierce, W. D. 1910. Some new species of weevils of economic importance. Journ. Econ. Ent. 3: 356–366.
———, 1913. Miscellaneous contributions to the knowledge of the weevils of the families Attelabidae and Brachyrhinidae. Proc. U.S.N.M. 45: 365–426.
Say, T. 1831. Descriptions of North American Curculionides. New Harmony. 30 p.
Schönherr, C. J. 1834. Genera et species Curculionidum. II, 1, p. 1–326. Paris.
———. 1843. Genera et species Curculionidum. VII, 1, p. 1–479. Paris.
Seidlitz, G. von. 1865. Monographie der Curculioniden-Gattung *Peritelus* (Germ.). Berl. Ent. Zeit. 4: 271–355.
Van Dyke, E. C. 1936. New species of North American weevils in the family Curculionidae, subfamily Brachyrhininae. V. Pan-Pac. Ent. 12: 73–85.

Astronomy. *Freeman D. Miller.* Bellman Publishing Company, Cambridge, Mass. 1955. 32 pp. $1.00.

This booklet is No. 72 in the Vocational and Professional Monograph Series issued by this company. The author, a member of the Astronomy Department of the University of Michigan, presents in a concise manner the facts which would be of interest to anyone considering the profession of astronomy. After a short introduction concerning its historical development, the status of modern astronomy is briefly discussed. The nature of the work of the modern astronomer is outlined, and the personal qualifications desirable in this profession are listed. The nature and the cost of training in the field are discussed in rather specific terms, and the employment opportunities indicated.

This is the first of this series which has come to the attention of the reviewer, and if the other vocations and professions are treated as well as astronomy, these monographs should be available in all high school, college, and university libraries, to provide valuable guidance to our students.

NICHOLAS MOGENDORFF

DETERMINATION OF CALCIUM IN CINDER BLOCK

ARTHUR H. BLACK AND VANCE H. DODSON
Department of Chemistry, University of Toledo, Toledo, Ohio

The use of Versene (ethylenediaminetetraacetic acid or EDTA) for the determination of calcium ion in aqueous solution was introduced by Schwarzenbach and coworkers in the years 1946 to 1948. Since then numerous papers have been published on the determination of calcium in various mixtures using this reagent. The measurement of the calcium content of limestone has been reported by several experimenters (Cheng *et al.*, 1952, 1953; Gehrke *et al.*, 1954). Analytical procedures for the determination of calcium in soil and plant products (Cheng *et al.*, 1951) and in water (Love, 1953; Diehl *et al.*, 1950) have also been outlined in the literature.

In the study of the physical properties of cinder block, currently being made at the University of Toledo, the calcium oxide and silica present, as complex silicates, has been of considerable interest. The recommended procedure for the analysis of cement products calls for a gravimetric determination of the silica and calcium oxide (Scott, 1939). These methods are very slow and time consuming. In an effort to simplify the analysis, a shorter volumetric method was investigated for the determination of the calcium employing Versene as the titrant.

GRAVIMETRIC PROCEDURE

The gravimetric procedure used in the determination of the acid soluble calcium silicates in cinder block is essentially the same as that outlined by Scott.

1. Representative samples of the block to be analyzed are crushed to pass a no. 20 screen, quartered, and further crushed to pass a no. 70 screen. A 10 g. sample of the finely divided material is heated with 250 ml. of water and 50 ml. of hydrochloric acid (sp. gr. 1.19) and allowed to boil for three minutes with continuous stirring. The mixture is diluted with water to a volume of 400 ml. and digested on a steam bath for 15 min. The mixture is then filtered while hot and washed with hot water.
2. The filtrate is diluted to 500 ml. and two 100 ml. aliquots are withdrawn for the gravimetric analysis of silica and calcium oxide.
3. The silicon dioxide is removed from the sample solutions after a double dehydration process.
4. The aluminum and iron oxides are removed by a double precipitation process.
5. Finally, the calcium is precipitated as the oxalate by the double precipitation method and ignited (at 1200°C) to calcium oxide in platinum crucibles.

VERSENATE METHOD

In the Versenate method, used by the authors for the determination of calcium oxide in cinder block, Step 1 (of the previously described gravimetric procedure) is carried out and the filtrate is diluted to 500 ml. (Step 2). Two 100 ml. aliquots are withdrawn and set aside for the silica determination (Step 3) and three 10 ml. portions of the remaining filtrate are taken for the calcium determinations, using Versene.

Reagents. Versenate Solution—4 g. of the analytical grade di-sodium Versenate, furnished by Versenes Incorporated, are dissolved in recently boiled and cooled distilled water and diluted to 1 liter.

Standard Calcium Solution—1 g. of analytical grade, low magnesium content, calcium carbonate is dissolved in a small amount of dilute hydrochloric acid and diluted to 1 liter. (1 ml. of standard solution = 0.5608 mg. calcium oxide).

Potassium Hydroxide—10 percent aqueous solution of analytical grade potassium hydroxide.

Murexide Indicator—a 0.2 g. portion of Eastman Kodak's Murexide thoroughly ground together with 40 g. of analytical grade potassium sulfate.

Triethanolamine—10 percent aqueous solution of U. S. P. triethanolamine.

Standardization of the Versenate Solution. A 25.0 ml. aliquot of the standard calcium solution is transferred to a 200 ml. porcelain casserole. Ten drops of the triethanolamine solution are added, and the pH of the solution is raised to 12.5 (measured with pHydrion paper) by the careful addition of the 10 percent potassium hydroxide. Approximately 40 mg. of the indicator powder is added by means of a small scoop. The Versenate solution is then run into the casserole from a burette until the color of the indicator changes from pink to a permanent violet.

TABLE 1

Determination of Calcium in Standard Calcium Carbonate Samples Using Versene

Sample Number	Calcium Oxide %	
	Reported Value	Versenate Method
1	8.01	7.96
2	16.93	17.01

TABLE 2

Determination of Calcium in Cinder Block

Sample Number	Calcium Oxide %		Difference
	Gravimetric Method[a]	Versenate Method[b]	
1	7.59	7.55	−0.04
2	7.57	7.46	−0.11
3	7.71	7.75	+0.04
4	17.35	17.20	−0.15
5	7.52	7.45	−0.07
6	7.24	7.33	+0.09
7	16.93	16.84	−0.09
8	17.65	17.48	−0.17
9	6.64	6.50	−0.14

[a] Average of duplicates.
[b] Average of triplicates.

Procedure for the Analysis of Cinder Block by Versene. Each of the 10 ml. aliquots is transferred to a 200 ml. porcelain casserole, diluted to about 30 ml. and treated in the manner described for the standard calcium solution. For samples containing large amounts of iron it is advisable to add at least 16 to 20 drops of the triethanolamine solution.

RESULTS OF ANALYSIS

Before the standard Versenate solution was used in the analysis of the cinder block, the calcium content of two samples of calcium carbonate, which had been

previously analyzed by standard gravimetric and volumetric methods (Diehl, 1952) was determined by the proposed procedure. The samples were dissolved in dilute hydrochloric acid and treated as outlined in the standardization of the Versenate solution. The results are reported as percent calcium oxide (table 1). To ascertain the accuracy of their technique and the sensitivity of the method, the authors felt that a check should be made on samples of known calcium content.

A comparison of the calcium oxide content of cinder block, as determined by the gravimetric method and by the Versenate method, is shown in table 2.

CONCLUSION

The differences shown in table 2 between the calcium oxide content of cinder block measured by the two aforementioned methods are of little significance. This is especially true when one considers the non-homogeneity of the material being analyzed. Differences as high as 1 percent in calcium oxide content have been noted in samples taken from opposite ends of a given cinder block.

The use of the Versenate method, outlined here-in, is well justified both from the accuracy attained and the time and labor saved. The method does not require the removal of silica and the R_2O_3 group prior to the determination of calcium. Further, there is no need to separate magnesium by the double precipitation of the calcium as the oxalate. In short, the Versenate method enables one to determine the total calcium content of the acid soluble material in cinder block within a few minutes after solution of the sample.

ACKNOWLEDGMENT

The authors express their appreciation to Professors E. L. Saxer and G. L. Kalousek, of the University of Toledo, for supplying the cinder block samples.

LITERATURE CITED

Biedermann, W., and **G. Schwarzenbach.** 1948. Titration of alkaline earth metals using Eriochrome Black T as indicator. Chimia (Prague) 2:56.

Cheng. K. L., and **R. H. Bray.** 1951. Determination of magnesium and calcium in soil and plant material. Soil Sci. 72: 449.

Cheng, K. L., T. Kurtz, and **R. H. Bray.** 1952. Determination of calcium, magnesium and iron in limestone. Anal. Chem. 24: 1640.

———, **R. H. Bray,** and **T. Kurtz.** 1953. Determination of total iron in soils by disodium dihydrogen ethylenediamine tetraacetate titration. Anal. Chem. 25: 347.

Diehl, H., C. A. Goetz, and **C. G. Hach.** 1950. Determination of water hardness. J. Am. Water Works Assoc. 42: 33.

———, and **G. F. Smith.** 1952. Quantitative Analysis. John Wiley and Sons, New York.

Gehrke, C. W., H. E. Affsprung, and **Y. C. Lee.** 1954. Direct ethylenediamine tetraacetate titration methods for magnesium and calcium. Anal. Chem. 26: 1944.

Love, S. K., and **L. L. Thatcher.** 1952. Review on water analysis. Anal. Chem. 24: 296.

Schwarzenbach, G., and **H. Ackermann.** 1947. Ethylenediamine and chloroacetic acid. Helv. Chim. Acta. 30: 1798.

———, **W. Biedermann,** and **F. Bangerter.** 1946. Determination of water hardness. Helv. Chim. Acta. 29: 811.

Scott, W. W. 1939. Standard Methods of Chemical Analysis. Vol. 2, Ed. 5. D. Van Nostrand Co., New York.

Petrographic Mineralogy. *Ernest E. Wahlstrom.* John Wiley & Sons, New York. 1955. vii + 408 pp. $7.75.

Important additions, particularly extensions of the treatment to include sedimentary and metamorphic rocks, justify the change in title from IGNEOUS MINERALS AND ROCKS, which appeared in 1947. Usefulness as a textbook for a general course in petrography is enhanced through the revision and enlargement.

DUNCAN MCCONNELL

OXYGEN CONSUMPTION OF ADRENALECTOMIZED AND RENAL DAMAGED FROGS[1,2]

CLIFFORD A. ANGERER AND MARY C. MURRAY

Department of Physiology, College of Medicine, The Ohio State University, Columbus 10

The frog, both as a representative of poikilotherms and as experimental material for adrenocortical investigations, possesses certain advantages over homoiotherms for studies involving penetration rates of known molecular-volume vital dyes and of radioactive physiological ions through its integument (Angerer, 1942), of water into and from the whole organism (Angerer and Angerer, 1949; Angerer, 1950) and into and from skeletal muscles (Angerer and Angerer, 1950), and for studies treating of skin bio-electric potentials (Williams and Angerer, 1950). Recently success has been attained with injections of adrenocortical extract (ACE) into otherwise normal frogs with regard to metabolism (Calhoon and Angerer, 1951) and skin bio-electric potentials (Williams and Angerer, 1952). In the light of these studies and of the paucity of data treating of the metabolism of normal frogs, it was considered desirable (a) to determine for the first time the rates of oxygen consumption at various intervals of time after renal damage and (b) after adrenalectomy, with the view of extending the study of adrenocortical function to the poikilotherms, and (c) to obtain data for the oxygen consumption of another species of frogs by a different respirometric technique.

METHOD

Unfed male frogs, *Rana pipiens*, of winter (late autumn and winter) and early spring (before spawning) stock, weighing between 20 and 30 gm. and showing testicular fat at autopsy, were divided into 3 major groups (12–26 frogs group): (1) adrenalectomized, both "paired" (curve A', fig. 1) and "unpaired" (curve A), (2) renal damaged (sham-operated control, curve R), and (3) unoperated (normal control, curve N) frogs. The method of adrenalectomy, of renal damage, of postoperative care on sphagnum moss, and of determining adrenocortical insufficiency has been described elsewhere (Angerer and Angerer, 1949; Angerer, 1950). "Paired" animals (included in groups 1 and 2) served as their own controls prior to adrenalectomy; "unpaired" frogs had their controls (unoperated and renal damage) randomly and simultaneously selected from the same stock as those designated for a given operative procedure.

Oxygen consumption was determined by direct-reading volumeters. The accuracy of this technique is comparable to that of the Winkler method (Calhoon and Angerer, 1950, 1953). The frogs were gradually acclimated (17–21 hr.) to the temperature of the waterbath to prevent possible thermal stimulation (Angerer, 1940). The frogs were permitted to sit in a given volume of tapwater sufficient to moisten their integuments. The volumeters were oscillated through a distance of 3 cm., at a frequency of 110 cycles/min. and at a bath temperature of 30 ± 0.01° C. After a 15 minute equilibration period, readings were made immediately at 15-minute intervals for at least one hour. All readings were corrected to standard pressure and unit body weight. The results were expressed as ml. oxygen consumed/gm. of body weight (wet)/hr.

[1]Aided by grants from the Comly-Coleman Research Fund, College of Medicine, and from the Graduate School, The Ohio State University.

[2]Presented at the Columbus meetings, American Physiological Society: Amer. Jour. Physiol. 163: 737. 1950.

RESULTS AND DISCUSSION

Normal frogs. The mean oxygen consumption for all (38) frogs studied is 0.175 ± 0.003 (standard error) ml./gm./hr. at 30° C. There is little fluctuation in this mean over 5 successive days as is indicated by the low value for its standard error. Curve N, figure 1, serves not only for purpose of reference when the percent

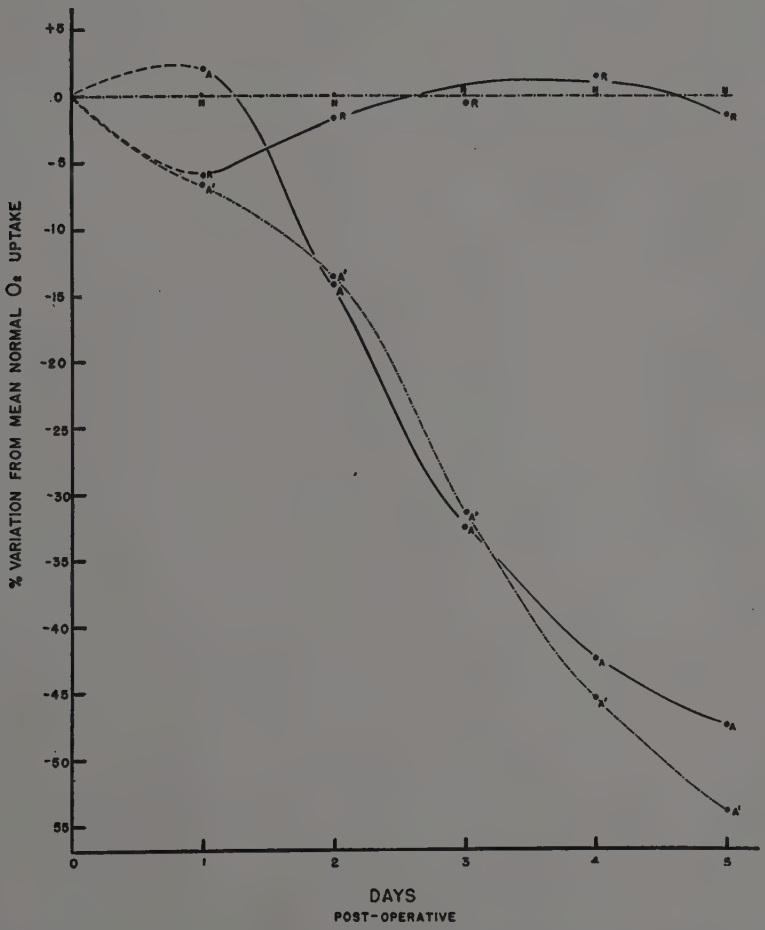


FIGURE 1. The percent variation in mean oxygen uptake (ml./gm./hr.) for renal damaged (R) and for "paired" (A') and "unpaired" (A) adrenalectomized frogs compared with normal (N) frogs (ordinates) is plotted against their postoperative time in days (abscissae). "Basal" winter and spring frogs were studied at 30° C. ("Paired" indicates each frog served as its own control.)

variation in mean oxygen uptake (ordinate) is plotted against postoperative time in days (abscissa) but also indicates, incidentally, the mean oxygen uptake (0.175 ml./gm./hr.) of all normal frogs during a period of 5 successive days. When winter and spring frogs are studied, under the stated experimental conditions,

the means are 0.183 ± 0.006 and 0.165 ± 0.006 ml. O_2/gm./hr., respectively. The difference (−9.8%) between these values is not significant ($P > 0.05$, Student's method); therefore, the mean for all stocks is used in subsequent comparisons. The lack of significance between the means for our winter and spring stocks is probably due to all frogs being kept under routine laboratory conditions, as previously outlined, and in a "basal" state, in that the frogs were unfed and subsisted on their fat depots. The values hitherto reported (Loewy, 1926) for 2 frog species, studied at 20° C, are 0.080 and 0.260 ml. O_2/gm./hr. for winter and April *R. temporia* and 0.100 and 0.210 ml. O_2/gm./hr. for winter and April *R. fusca*, respectively. Our mean of 0.175 ml. O_2/gm./hr. at 30° C, when corrected to 20° C, on the assumption that the Q_{10} is *ca.* 2.3, gives a value identical with that which is found for winter *R. temporia*. April frogs, to be sure, would have a higher metabolic rate due to their feeding, increased physical activity and incipient spawning. Indeed, Krogh (1904) found the highest oxygen uptake occurs during the spawning season. Krogh was interested in another phase of frog metabolism and, owing to experimental necessity, forcibly confined his animals. Even so, he obtained values of *ca.* 0.108 and 0.59–0.113 ml. O^2/gm./hr. at 20° C for winter *R. fusca* and *R. esculenta*, respectively.

It is interesting to note that Calhoon and Angerer (1955) using the same apparatus, frog stock and experimental technique, except for the omission of shaking of the volumeters during the course of the experiment, (cf. their results, 1953, p. 242) obtained a mean of 0.156 ml. O^2/gm./hr. at 30° C. This may represent a truer "basal" value for *R. pipiens*, though the value is approximately at the limit of experimental error and its statistical significance is questioned.

Renal damaged frogs. The daily mean values for the oxygen consumption of 12 renal damaged frogs is presented in curve R, figure 1. After an initial decrease of 6.8 percent at the end of the first postoperative day, the data show little variation thereafter with time when compared with normal frogs (curve N). There is no statistical significance between these values (curve R) and those for normal frogs (curve N). All subsequent comparisons, unless otherwise stated, are made with unoperated frogs. These controls are used in place of renal damaged frogs in order that data from "paired" and "unpaired" adrenalectomized frogs may be compared. There is little reason to believe that sham operations with their concomitant renal damage would alter the oxygen uptake of amphibious and aquatic animals as long as the regenerative area is relatively small compared to the total respiring tissue mass. Furthermore, the amount of renal damage arising, of necessity, as a result of adrenal ablation, is apparently without injurious effects to the frog (Angerer, 1950).

Adrenalectomized frogs. Figure 1 gives data for the daily mean rates of oxygen consumption for "paired" (curve A') and "unpaired" (curve A) adrenalectomized frogs compared with their controls (curve N). While 71 percent of the "paired" frogs show a decrease in mean oxygen consumption at the end of the first postoperative day, a statistically significant decrease (*ca.* 13%, $P < 0.05$) is observed by the end of the second postoperative day. Thereafter the values decrease until the end of the fifth day when a mean of 0.095 ml. gm. hr. is obtained. This value is *ca.* 49 percent below that for the mean initial (zero) time or for the renal damaged frogs at the end of a corresponding postoperative interval.

A comparison of data for "paired" (curve A') and "unpaired" (curve A) adrenalectomized frogs shows that both sets of data fall along essentially sigmoid curves. Whether the curve form is significant, or fortuitous, is problematical. At best, it suggests many reactions are occurring simultaneously. To our knowledge a comparable treatment of data for warm blooded animals does not exist. Secondly, there is a consistently greater decrease in mean oxygen uptake by the end of the fifth postoperative day for both winter (10%) and spring (12%) "paired" than for "unpaired" adrenalectomized frogs. Little variation is observed between

winter (0.101 ml. 0_2/gm./hr. and spring 0.089 ml.) "unpaired" adrenalectomized frogs by the end of the fifth postoperative day. Thirdly, the death point for a majority (*ca.* 65%) of the adrenalectomized frogs under prevailing experimental conditions occurs by the end of the fifth day, whereas normally it is between the seventh and twelfth postoperative day (Angerer, 1950). The last two items are probably to be interpreted as due to the stress induced in handling and in subjecting the frogs to the relatively high environmental temperature. These two stressing agents would increase the demand for unavailable adrenocortical hormone(s) (Hartman and Brownell, 1949). A decrease in metabolism following adrenalectomy has been noted for several homoiotherms: cats (Aub, Forman and Bright, 1922), rabbits (Marine and Baumann, 1922), rats (Carr and Beck, 1927), and so on. The chief difference is the extent of the metabolic depression between homoiotherms (*ca.* 25%) and poikilotherms (*ca.* 50%).

SUMMARY

The oxygen consumption of 20–30 gm. male frogs, *R. pipiens*, was studied by direct-reading volumeters. All frogs studied may be placed in 3 major groups: *A*, unoperated (normal control, curve N, fig. 1); *B*, renal damaged (sham operated control curve R); *C*, "paired" (curve A') and "unpaired" (curve A) adrenalectomized frogs (see "method").

1. The mean oxygen consumption for winter and spring frogs under "basal" conditions is 0.175 ± 0.003 ml./gm./hr. at 30° C; no statistical difference is observed between winter and spring stocks under the experimental conditions employed.

2. There is no significant difference between the mean values for oxygen uptake for excessive renal damaged and normal frogs (cf. curves R and N).

3. Adrenalectomized frogs show no significant change in oxygen consumption at the end of the first postoperative day compared with unoperated or renal damaged frogs. Thereafter, the means for both "paired" (curve A') and "unpaired" (curve A) adrenalectomized frogs fall along a sigmoid curve (fig. 1). On the fifth postoperative day a mean of 0.095 ml. 0_2/gm./hr. is obtained. The figure represents a decrease of 46 percent below that of the controls.

4. The stress of repeated handling and of subjecting adrenalectomized frogs to relatively high environmental temperatures depresses both their metabolism and viability (cf. curves A' and A).

ACKNOWLEDGMENTS

We wish to thank Dr. Thomas B. Calhoon, formerly of this laboratory and now of the Department of Physiology, Medical College of South Carolina, Charleston, for his help to M. C. M. in conjunction with the use of the respirometers.

REFERENCES

Angerer, C. A. 1940. The effect of thermal increments on and the subsequent adjustment of the protoplasmic viscosity of *Amoeba proteus*. Physiol. Zool. 13: 219–226.

———. 1942. Unpublished data.

———. 1950. Body weight, survival time, coloration, and water content of skeletal muscles of adrenalectomized frogs. Ohio Jour. Sci. 50: 103–106.

———, and **H. H. Angerer.** 1949. The rate and total loss of body water on the survival time of adrenalectomized frogs. Proc. Soc. Exper. Biol. and Med. 71: 661–665.

———, and ———. 1950. Weight variations of adrenal ectomized frog muscles in different concentrations of Ringer's solutions. Proc. Soc. Exper. Biol. and Med. 73: 265–268.

Aub., J. C., J. Forman, and **E. M. Bright.** 1922. The effect of adrenalectomy upon the total metabolism of the cat. Amer. Jour. Physiol. 61: 326–348.

Calhoon, T. B., and **C. A. Angerer.** 1950. The effect of relatively large doses of sesame oil on oxygen consumption and the antagonistic action of adrenocortical extract. Endocrinol. 46: 327–333.

———. 1951. Use of a new respirometer in studying the effects of adrenocortical extract on total metabolism. Fed. Proc. 10: 23.
———. 1953. A volumeter for respiration of aquatic animals. Ohio Jour. Sci. 53: 240–243.
———. 1955. Adrenal cortical extract on the oxygen consumption of normal frogs. Physiol. Zool. In press.
Carr, C. J., and **F. F. Beck.** 1937. The metabolism of adrenalectomized rats. Amer. Jour. Physiol. 119: 589–592.
Krogh, A. 1904. On the cutaneous and pulmonary respiration of the frog. (A contribution to the theory of the gas exchange between the blood and the atmosphere.) Skand. Arch. Physiol. 15: 328–419.
Hartman, F. A., and **K. A. Brownell.** 1949. The adrenal gland. Lea and Febiger. Philadelphia.
Loewy, 1926. Gas-und energie-wechsel. Tabulae Biologicae Periodicae 3: 504.
Marine, D., and **E. J. Baumann.** 1922. VI. Further data on the effect of suprarenal insufficiency (by removal) in rabbits. Jour. Metabol. Research 2: 1–18.
Williams, M. W., and **C. A. Angerer.** 1950. Adrenalectomy on frog skin potentials. Fed. Proc. 9: 136.
———. 1952. Unpublished data.

Animal Agents and Vectors of Human Disease. *Ernest Carroll Faust.* Lea and Febiger, Philadelphia. 1955. 660 pages. $9.75.

Lea and Febiger published Dr. Faust's *Human Helminthology* and *Clinical Parasitology* so it is not surprising that this new book is like these well known textbooks in format. Also many of the illustrations are the same in all three volumes.

The features of *Animal Agents and Vectors of Human Disease* which make it a distinctive contribution to the science of parasitology are its broad coverage, moderate length, condensation of much information into handy tables, excellent chapter summaries, a section on technical aids, and most important of all a point of view indicated in the book's title, and elaborated in the introductory chapters, that often fails to develop sufficiently in many teachers and their students of parasitology. The illustrations are excellent and many are new. The absence of color on the seven plates of malarial parasites fortunately will force the user to give attention to the morphology of erythrocytes and parasites.

There are eight sections as follows: 1. General information and orientation, 2. Protozan agents of disease, 3. Roundworms, 4. Tapeworms and flukes, 5. Other helminths parasitizing man, 6. Arthropods as agents and vectors, 7. Other invertebrate and vertebrate agents of disease, and 8. Technical aids. This information will be useful to teachers, students, and the personnel of zoological, medical, and public health laboratories. The chapters have lists of pertinent literature. There is an index of authors and a subject index.

CARL VENARD

Applied Entomology. *H. T. Fernald* and *H. H. Shepard.* McGraw-Hill Book Co., Inc., New York. 5th edition, 1955. x + 385 pp. $7.00.

Following the format of previous editions, this revised fifth edition covers only a few pest species in each of the major families of the commonly recognized orders of insects. The presentation is from the classification or systematic viewpoint.

A large portion of the text has been rewritten. Some species of lesser importance have been dropped, others of greater importance have been added. From the standpoint of applied entomology there are still some important pest species not covered. What to exclude and what to include is always a problem for the textbook writer and the purpose for which the book was intended seems to be the deciding factor. The control measures given for many of the pests are not the most recent recommendations. However, in most instances they will give the reader a satisfactory solution to his pest control problem. Brief mention of more of the recently adopted chemicals would have been a welcome addition and would not have enlarged the book greatly. Slight changes in the arrangement of the chapters have been made to conform to new ideas on classification. Near relatives of insects are given more consideration. Numerous illustrations have been added and others deleted. A few of the new ones do not appear as good as similar ones in the previous edition. Numerous footnotes have been added showing the pronunciation of orders, families, suborders, subfamilies, and other technical terms.

In spite of the shortcomings mentioned the book should satisfactorily serve as a textbook for general or introductory courses in entomology.

R. H. DAVIDSON

MULTICELLULAR ROOT HAIRS ON ADVENTITIOUS ROOTS OF *KALANCHOE FEDTSCHENKOI*[1]

RICHARD A. POPHAM AND ROBERT D. HENRY

Department of Botany and Plant Pathology, The Ohio State University, Columbus 10

In the course of examining root tips of various greenhouse plants, freehand sections were made of the stiff, pink-colored aerial adventitious roots of *Kalanchoe fedtschenkoi*. Upon examining sections of these roots, structures resembling multicellular root hairs were observed. Since we were unable to discover an account of multicellular root hairs in recent botanical literature or a textbook of botany, we decided to look into the matter further.

Dittmer (1949) states that he has never seen a septate root hair although he examined "many millions of root hairs on hundreds of plants". He does mention that some of the earlier workers reported septate root hairs but that the structures described were judged by "many workers since" to be fungal hyphae. A rather thorough survey of the literature failed to yield the name of a single specific plant for which multicellular root hairs are claimed.

We at first speculated that the structures which superfically looked very much like aerial adventitious roots of *Kalanchoe* might in reality be abnormal leafless branches with trichomes. However, if they turned out to be adventitious roots we surmised that they might possibly have arisen exogenously as do the adventitious roots of *Radicula aquatica*, in which case the hairs might also be out-growths of cells of the epidermis of the stem. In water cress, epidermal cells of the stem in the vicinity of the exogenous root primordium become meristematic and form a continuous layer of epidermis over the entire root primordium. Both of these possibilities were investigated even though we found no epidermal hairs of any kind on the smooth stems of *Kalanchoe fedtschenkoi*.

MATERIAL AND METHODS

Adventitious root primordia and many sizes of adventitious roots including branched roots seven or more inches long were studied. Fresh freehand sections as well as microtomed transverse and longitudinal sections were prepared. Pieces of aerial adventitious roots preserved in FPA (5 ml. formalin, 5 ml. propionic acid, 90 ml. 50 percent ethyl alcohol) were dehydrated in an ethyl alcohol series and embedded in a special paraffin embedding mass (Popham, 1947) using toluene as the vehicle for infiltration. Microtomed sections were cut at 10 μ and were stained with safranin and anilin blue according to a procedure described by Popham *et al.* (1948).

ROOT APEX ZONATION

An examination of transverse sections of the aerial adventitious roots revealed a rather large pith, xylem (usually 10 to 15 areas) and phloem in radial polyarch arrangement, a pericycle of 1 to 3 cell layers, a uniseriate endodermis, 3 to 5 layers of cortical parenchyma, a uniseriate hypodermis which is poorly differentiated from cells of the cortical parenchyma, and a uniseriate epidermis. Neither stelar cambium nor cambium initials were observed. Anthocyanin was clearly visible in cells of the cortex of fresh, freehand transverse sections.

[1]Papers from the Department of Botany and Plant Pathology, No. 581. Supported in part from funds allocated by the University Advisory Committee on Research Grants.

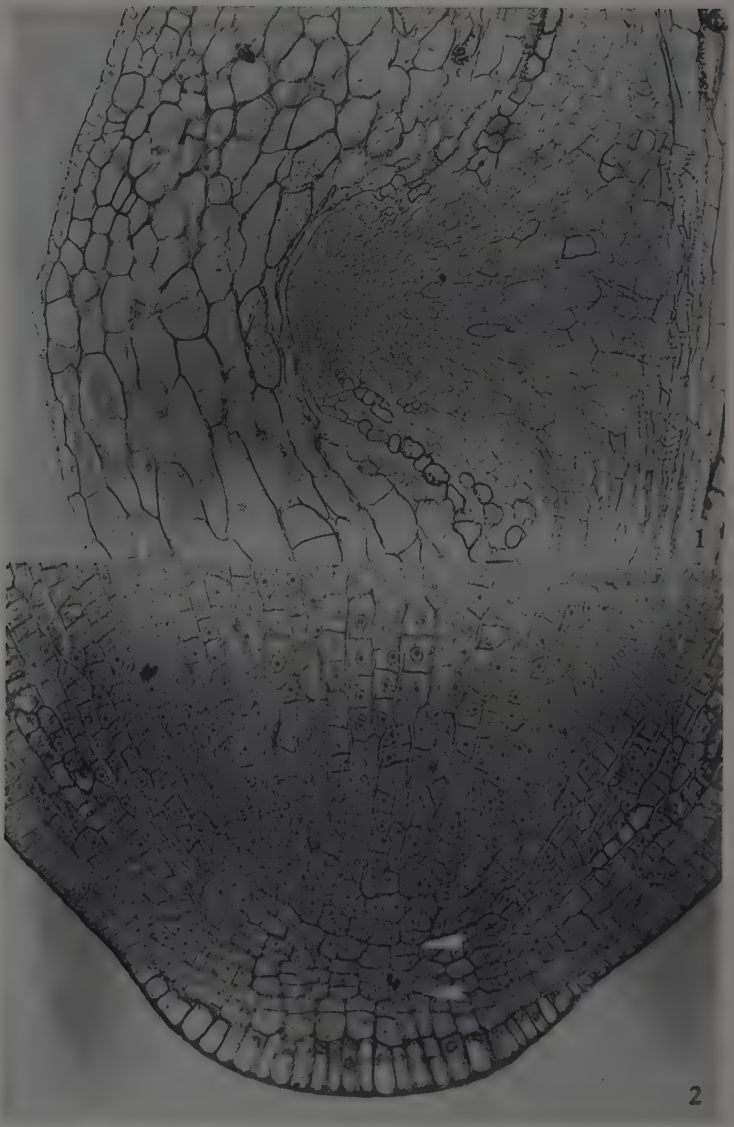


FIGURE 1. Longitudinal section of stem and aerial adventitious root illustrating the endogenous origin of the latter. X120.

FIGURE 2. Median longitudinal section of aerial adventitious root tip illustrating Janczewski's type III, *Convolvulus* sub-type, of zonal organization in the promeristem. The plerome lies just above the top indicator marker, the several-layered periblem lies between the two markers, and the dermato-calyptrogen lies just below the lower marker. X330.

In median longidutinal sections of the apex we found an unmistakable root apex organization (fig. 2). Three histogens[2] were observed: (1) a plerome, (2) a periblem, and (3) a dermato-calyptrogen. This zonal pattern is, in general, the same as that frequently designated as the common dicotyledenous type, Janczewski's (1874) type III. However, the cortex of *Kalanchoe fedtschenkoi* originates from two or more (usually more) layers of cells (periblem) lying between the plerome and the dermato-calyptrogen (fig. 2). The hypodermis does not originate from a separate histogen.

FIGURE 3. Longitudinal section of aerial adventitious root tip with multicellular root hairs within 380 μ of the tip cell of the root. Notice that a root hair in an early stage of formation is farther from the tip than those composed of many cells. X130.

In describing his type III zonation of roots, Janczewski states that the cortex of *Helianthus* and *Fagopyrum* originates from one or more cells disposed in a *single* layer (periblem). He regarded the root promeristems of *Linum usitatissimum* as representing a sub-type of this general zonal organization. In *Linum* the cortex originates from two layers of cells, the outer of which gives rise to the hypodermis exclusively. Although the general zonal pattern is the same for *Kalanchoe* as for *Helianthus* and *Linum*, the multi-layered condition of the periblem

[2]Some anatomists seem to feel that the use of Hanstein's histogen terminology and Janczewski's typology imply a constancy of root apex zonation for the individual plant and/or the species. Be this as it may, we have used their terms solely as a convenient means of describing what we observed.

places the root of *Kalanchoe* in a separate and second sub-category of Janczewski's type III.

Just such a sub-category was described by Eriksson (1878) and was named the *Convolvulus* sub-type. Janczewski's type III and his *Linum* sub-type were recognized by Eriksson (1878) although he referred to them as type I and the *Linum* sub-type. The *Convolvulus* sub-type, which Eriksson discovered, is characterized by a periblem consisting of three to several superposed layers of cells. The cells of the outer layer are described as dividing anticlinally and only occasionally periclinally. Root apices of the following plants were found by him to exhibit this *Convolvulus* sub-type of zonal organization: *Hoja carnosa* (*Asclepiadeae*); *Abutilon molle*, *Pavonia weldini*, *P. spinifex* (*Malvaceae*); *Villarsia nymphaeoides* (*Menyantheae*); *Convolvulus cneorum* (*Convolvulaceae*); *Begonia nelumbifolia*, *B. ricinifolia*, *B. glacialis* (*Begoniaceae*).

Neither Janczewski (1874) nor Eriksson (1878) recorded observations on a species belonging to the *Crassulaceae*. Adventitious root primordia of *Sedum sieboldtii* and *Bryophyllium calycinum* as well as lateral root primordia of *Sedum acre* and *S. maximowiczii* were studied by Van Tieghem and Douliot (1888). They described them as having the common dicotyledenous type of root zonation with a periblem composed of a single cell or a single layer of cells. The same type of organization was seen by Koch (1874–77) in root apices of *Sedum spurium* and *S. album*. In the root apex of another plant (*Umbilicus horizontalis*) belonging to the *Crassulaceae*, Flahault (1878) observed and described a zonal organization like Janczewski's type III. The periblem in the promeristem of *Umbilicus* is a single layer of cells.

We were unable to discover a description in the literature which claimed Eriksson's (1878) *Convolvulus* type of zonal organization for the root apex of any species belonging to the family *Crassulaceae*.

ORIGIN AND GROWTH OF THE MULTICELLULAR ROOT HAIRS

After thus completely satisfying ourselves that the structures observed on *Kalanchoe* aerial stems were roots, we turned our attention to the problem of ascertaining whether the root hairs arose from a bonafide root epidermis or whether they arose from cells of stem epidermis overlying the root apex proper.

Serial transverse and longitudinal sections were made of portions of stems on the surface of which slight protuberances were observed. After sectioning many such stem pieces, we were able to assemble an intergrading series of longitudinal sections of adventitious root primordia and adventitious roots whose apices had grown several millimeters beyond the epidermis of the stem. All of these sections showed evidence of an endogenous root origin (fig. 1). The adventitious roots probably arise in phloem parenchyma although this point was not surely confirmed. In any event, the origin is internal to the cortex since the layers of cortex are compressed and subsequently punctured as the root primordium enlarges and elongates. In every case where the root apex was found protruding beyond the epidermis of the stem, the epidermis was clearly broken and showed no evidence of continuity with that of the root.

One of the facts that had originally made us doubtful as to whether we were observing bonafide root hairs was their occurrence at levels extremely close to the root tip, even on roots several centimeters long. Many were found as close as 380 μ back of the tip cell of the root cap (fig. 3); some were seen closer to the root tip. Many persisted as long as the epidermis remained firmly attached to the root but hairs composed of living cells were usually found less than 1 cm. from the root tip.

The first sign of root hair formation was the radial elongation of one of the typically radially elongated cells of the epidermis. Accompanying the radial elongation of the trichoblast was an expansion of the outer end of the cell and

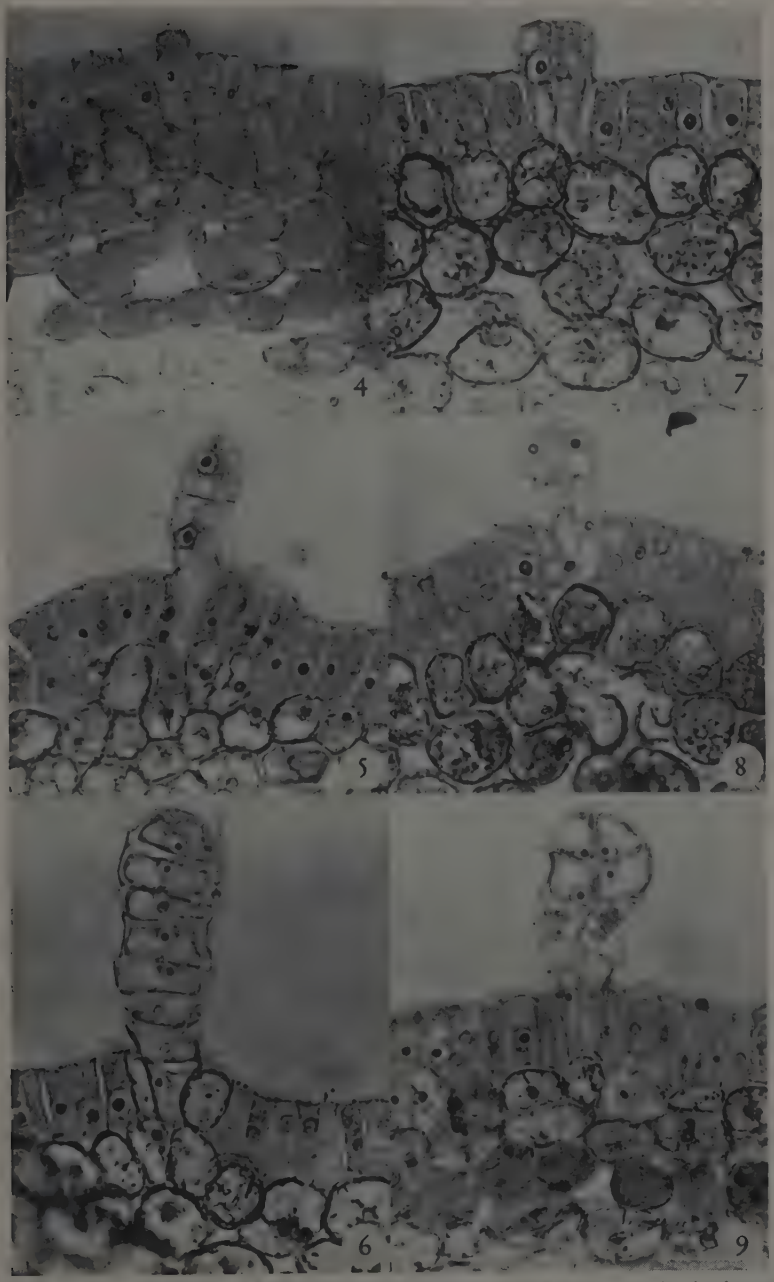


FIGURE 4–9. Cross sections of aerial adventitious roots illustrating stages in the formation of uniseriate and biseriate multicellular hairs. X650.

the contraction of the end of the cell adjacent to the hypodermis (fig. 4). During these early phases of root hair formation the nucleus retains an approximately central location in the cell (fig. 4, 7). In contrast to typical root hair formation during stages of which a *portion* of the free wall bulges, the *entire* free wall of the multicellular root hair initial was forced out into a dome. Concomitantly the nucleus moved outward toward the bowed surface of the cell. After becoming conspicuously more radially elongated and vacuolated than neighboring epidermal cells, the trichoblast divided either transversally (fig. 5) or longitudinally (fig. 7). In either event enlargement of the two cells formed was followed by one or several successive transverse divisions in the one (fig. 6) or the two (fig. 8 ,9) tiers of cells. Transverse wall formation in adjacent cells nearly always occurred at the same level and simultaneously resulting in two tiers of nearly equal size corresponding cells. It was inferred that growth of the hairs was due to divisions occurring in the terminal cell of each tier although confirming mitotic figures were not observed. On older portions of roots, hairs consisting of two tiers of cells sometimes split throughout one-fourth to three-fourths of their length. This appeared to be due to the dissolution of the middle lamellae holding cells of the two tiers together although no microchemical tests were conducted to verify this thesis.

Few hairs attain lengths greater than 75 μ. Due to the short maximum length and due to the fact that the maximum length is rapidly attained, zones composed of root hairs of decreasing lengths and ages, such as those often seen at root tips, are not evident in *Kalanchoe fedtschenkoi* (fig. 3).

HAIRS OF AERIAL VS. SUBTERRANEAN ADVENTITIOUS ROOTS

It appears somewhat incongruous that multicellular root hairs have not been found to occur on adventitious as well as on primary and lateral roots of plants having multicellular stem hairs. Most botanists will probably agree that anatomical form is controlled by and is the result of the combined influences of genes and the environment to which cells are exposed. If we wish to assume that the gene complements of cells of roots and stems of a plant are identical, differences in stem vs. root epidermal hair structure for any particular plant must be attributed to differences in the environment to which the cells are exposed. It would seem that occasionally, at least, epidermal cells of roots would be exposed to environmental conditions identical, or nearly so, to those of stem epidermal cells. It is therefore interesting that this first documented occurrence of multicellular root hairs is reported for *aerial* adventitious roots whose initiating epidermal cells are exposed to environmental conditions similar to those of stem epidermal cells. Hairs of the aerial roots of *Kalanchoe fedtschenkoi*, whether original adventitious roots or lateral roots arising from them, are multicellular. It is equally noteworthy, but not astonishing, that roots of the adventitious root system growing in the *soil* were found to have *unicellular* hairs. We did not observe multicellular hairs on any subterranean root, although we did not conduct an exhaustive search for them.

The adventitious root system of *Kalanchoe fedtschenkoi* would seem to afford experimental material for a new avenue of approach to the solution of a fundamental problem of living organisms, namely, what combination of environmental factors is responsible for the initiation of the process of mitosis?

SUMMARY

The zonal organization of the aerial adventitious root apex is described as conforming to Eriksson's *Convolvulus* sub-type of Janczewski's common dicotyledonous type III. A plerome, a several-layered periblem, and a dermatocalyptrogen are clearly defined in the root promeristem.

It was found that true multicellular root hairs arise from epidermal cells of aerial adventitious roots of *Kalanchoe fedtschenkoi*. The hairs were found to be uniseriate or biseriate. They persist until the epidermis separates from the root, although hairs composed of living cells are usually found only on the tip 1 cm. of the roots.

Hairs of adventitious roots growing in the soil are most commonly or always unicellular and look very much the same as hairs of many other roots.

It is suggested that the adventitious root system of *Kalanchoe fedtschenkoi* may provide suitable material for a new approach to investigations aimed at discovering the combination of environmental factors responsible for the initiation of mitosis.

Since completing this paper, we have found multicellular root hairs, similar to those described, on the aerial adventitious roots of *Kalanchoe somaliensis*, *K. daigremontiana*, and an unidentified *Kalanchoe* species.

In addition, two papers (Haberlandt, 1915 and Jurišić, 1934) dealing with glandular hairs ("Drüsenhaare") of roots were found. Diagrams accompany the former but not the latter paper. Descriptions of the hairs indicate that the structures which these authors saw on adventitious roots of several plants [*Bryophyllum pinnatum* and *B. crenatum* (Haberlandt, 1915); *Bryophyllum proliferum*, *B. verticillatum*, *B. daigremontianum*, and *B. pinnatum* (Jurišić, 1934)] are very similar to those described in this paper. These two papers have apparently been generally overlooked.

LITERATURE CITED

Dittmer, H. J. 1949. Root hair variations in plant species. Amer. Jour. Bot. 36: 152–155.

Eriksson, J. 1878. Ueber das Urmeristem-der Dikotylen—Wurzeln. Jahrb. Wiss. Bot. 11: 380–436.

Flahault, C. 1878. Recherches sur l'accroissement terminal de la racine chez les phanérogames. Ann. Sci. Nat. Bot. VI. 6: 1–168.

Haberlandt, G. 1915. Über Drüsenhaare an Wurzeln. Sitzungsber. d. Kgl. Preuss. Akad. d. Wiss., Phys.-Math. Kl. 1: 222–226.

Janczewski, E. de. 1874. Recherches sur l'accroissement terminal des racines dans les Phanérogames. Ann. Sci. Nat. Bot. V. 20: 162–201.

Jurišić, J. 1934. Zur Kenntnis der Drüsenhaare an den Wurzeln von *Bryophyllum*. Anzeiger der Akademie der Wissenschaften in Wien. 71: 192–195.

Koch, L. 1874–77. Untersuchungen über die Entwicklung der Crassulaceen. Verhandl. des Naturhistorisch—Medicinischen Vereins zu Heidelberg. 1: 421–442.

Popham, R. A. 1947. The importance of controlling cooling temperatures during embedding in paraffin. Science. 106: 475–476.

———, **T. J. Johnson,** and **A. P. Chan.** 1948. Safranin and anilin blue with Delafield's hematoxylin for staining cell walls in shoot apexes. Stain Tech. 23: 185–190.

Van Tieghem, P., and **H. Douliot.** 1888. Recherches comparatives sur l'origine des membres endogènes dans les plantes vasculaires. Ann. Sci. Nat. Bot. VII. 8: 1–660.

Plants Without Flowers. *Harold Bastin.* Philosophical Library, Inc. New York. 1955. xi + 146 pp. $6.00.

This book provides a popularized, but somewhat detailed account of the evolutionary theories, life histories, and economics of various groups of lower plants. A chapter is included on slime-fungi and bacteria, algae, lichens, true fungi, mosses, liverworts, ferns, fern allies, horsetails, and club mosses.

Examples are drawn chiefly from the flora of Great Britain, and common names are used in the text. However, a glossary of scientific names is included.

The book is well illustrated with photographs and line drawings, and is easy enjoyable reading.

DALE P. LILES

OBSERVATIONS ON THE HABITS OF TABANIDAE

R. L. BLICKLE[1]

Department of Entomology, University of New Hampshire, Durham

The following are observations made during the summers of 1952 and 1953 in the towns of Durham, Lee, Belmont, New Durham, and Gilmanton, New Hampshire. The observations are on hovering, flight activity, species frequenting flowers, and on species visiting moist areas on dirt roads. A method of moistening areas on dirt roads by artificial means and collecting flies visiting these areas is described.

HOVERING

Tabanus aurilimbus Stone; males were observed along dirt roads and woodland paths from July 6 to July 30. The insects hovered from 1 to 3 ft. above the ground surface and in each instance they were facing into the wind. The distance between individual flies varied from 6 ft. to over 150 ft. When another insect flew near the fly would make a "pass" at the intruder, return to the same location, and continue to hover. Flies otherwise disturbed would fly up or down the road for 50 to 100 ft. and return, in most instances, to the identical area. Undisturbed flies would hover in the same spot for several minutes.

The earliest observation was made at 9:00 A. M., the latest at 5:00 P. M., the hovering activity apparently being governed by light intensity and temperature.

Males of *T. aurilimbus* were very numerous in southeastern New Hampshire during the observation period. As many as 90 were noted on one day, July 13, 1952, in Durham, N. H.

There were no female horseflies taken or observed in conjunction with hovering males.

Tabanus trispilus Wiedemann; males of this species were seen to hover, momentarily, over pools or damp areas in dirt roads. These flies would dip once, or in some cases repeatedly, into the mud or water and dart away.

Tabanus actaeon Osten Sacken; this species was observed in the town of Durham, N. H., on August 31, 1951. The observers were Mr. W. J. Morse and Dr. O. T. Zimmerman. The insects were flying over and around houses and other buildings, there being no apparent pattern to the flight. The activity of these flies lasted one entire afternoon and has not been seen to occur since the above date. The observers estimated several hundred flies to be engaged in this activity. Of the dozen specimens taken, all were females.

ON FLOWERS

The following species were taken from various species of flowers. *Spireae latifolia*, meadow sweet, *Tabanus aurilimbus*, females; *T. superjumentarius*, female; *T. trispilus*, males and females; *T. typhus*, males and females; *Stonemyia rasa*, males and females; *S. tranquilla*, males and females; *Chrysops vittata*, female. From *Sium suave*, waterparsnip, males of *T. actaeon*, *T. quinquevittatus*, *T. trispilus*, *T. typhus*, males and females *S. rasa* and *S. tranquilla*. One male of *C. vittata* was taken from a corn tassel.

---

[1]Scientific Contribution No. 176 of the New Hampshire Agricultural Experiment Station.

AT LIGHTS

T. catenatus, females; *T. pumilus*, males; *S. rasa*, males; *C. geminata*, females; and *C. moecha*, females were taken at lights or in light traps.

MOIST AREAS ON DIRT ROADS

Twenty-four species, totaling 138 males and 114 females were taken from around puddles or moist areas in dirt roads in the towns of Durham, Gilmanton, and Lee, N. H.

The summer of 1952 was quite deficient in rainfall. After a rain shower on July 14 several tabanids were collected at a mud puddle. Whenever puddles were formed it was possible to collect tabanids from those areas. As the natural

TABLE 1

Tabanidae collected from dirt roads—1952–1953

Species	Dates	Male	Female
Tabanus actaeon	8/20 to 28	4	4
T. aurilimbus	7/14 to 25	6	1
T. catenatus	7/18 to 25	6	4
T. lasiopthalmus	7/14	..	1
T. lineola	6/29 to 7/21	13	16
T. nigrovittatus	7/25	5	1
T. nivosus	7/23	..	1
T. pumilus	6/26 to 7/27	9	8
T. quinquevittatus	7/18 to 25	1	2
T. sparus	7/26	..	1
T. sulcifrons	7/14 to 25	6	5
T. superjumentarius	6/26 to 7/27	20	20
T. trispilus	6/26 to 7/25	29	7
T. typhus	7/14	..	1
T. vivax	7/18 to 25	2	5
Stonemyia rasa	7/18 to 8/26	2	7
S. tranquilla	7/18 to 27	4	4
Chrysops aberrans	7/23	5	...
C. geminata	6/20 to 7/25	13	7
C. lateralis	7/10 to 25	10	6
C. montana	7/27	..	1
C. sackeni	7/9	..	1
C. striata	7/23 to 25	1	2
C. vittata	6/20 to 7/27	2	9
	Total.....	138	114

puddles were formed only at rare intervals, the following method was used with some success. Water was poured into depressions on dirt roads, especially in those depressions where natural puddles would form. A two gallon watering can of water was sufficient to treat an area. It was found that by keeping the moistened area small, about 3 ft. in diameter, it was easier to collect the flies. The location of the moistened area determined, to a great extent, the success of the method. Best results were obtained in partial shade adjacent to a bright open area. In one instance the use of an area in the open proved satisfactory; this occurred on a cool day when there was little air movement. Deep shade proved to be unsatisfactory.

The best collecting was done on days with bright sunshine, high temperature, and low humidity. Twelve species, 40 males and 21 females, were taken on

July 18, 1952. On this date the temperature was approximately 97° F, the humidity low, and the sky cloudless.

The flies, "cruising" along the road alight near the water or dip into the water. The larger tabanids usually drop to the muddy edges of the puddle and remain there for several minutes. The *Chrysops*, in almost every instance, will fly in, touch the mud or water, and fly off immediately. For this reason it was found that the *Chrysops* were more difficult to capture. Only one tabanid, *T. trispilus*, would hover over the water or mud and dip into the moistened area repeatedly. The other tabanids would halt momentarily over the area, dip once, and fly off immediately, or alight as mentioned above.

In capturing the flies the following method worked quite well. After the fly had alighted on the road the net was slowly placed over it, care being taken not to startle the insect. The closed end of the net was held extended, above the insect.The open end of the net was then placed quickly onto the ground, the fly being near the center of the open end. In nearly every case the tabanid would then fly up into the closed end of the the net. By a rapid sweeping motion the insect was entrapped. In rare instances the fly would attempt to crawl out from under the net rather than fly upward. If the ground was not too muddy, the closed end of the net was, in some cases, dropped onto the fly and the specimen picked up after it had become enmeshed. The method used is a matter of preference to the collector. Those insects which hovered over the moistened areas, and also in the case of the active *Chrysops*, were collected by rapid sweeping.

The use of the "watering can" method may work in periods of wet weather. In one area tabanids were taken from artifically made puddles, even though there was an abundance of water in a swamp on either side of the road. However, this method should prove more satisfactory in dry areas or in years of low rainfall.

Table I shows the number of specimens taken at various locations by the "watering can" method. Collecting was done for 3 days in June, 14 days in July, and for 3 days in August. Three species, *Tabanus sparus*, *Chrysops montana* and *C. sackeni* taken in 1953 were not taken in 1952. The earliest and latest dates of capture are given.

Handbook for Teachers of Earth Science. *M. F. Marple* and *W. C. Brown*. Information Circular No. 15, Ohio Division of Geological Survey, Columbus, 1955. vi + 61 pp., illus., maps. $0.50.

The publication is exactly what the title indicates—a handbook, which should be of great use to the teacher who wishes to explore with his class the inorganic natural world around them. Simple basic methods of exploration, collection, study, and identification are given in the principal fields of Earth Science—minerals, rocks, and fossils. In addition the study of the realted problems of water supply and soils are made meaningful, and a discussion of our mineral resources is included. An excellent short geologic history of the state is supplied. Many problems and projects are suggested which cover many levels of instruction, and all parts of the state. In fact the handbook might well be used as a text at some levels. Reference lists for books and sources of help are included. One for materials might well have been added.

Some indication of scale on the fossil drawings would be useful also. Perhaps the most serious fault found is the very limited space devoted to rocks, especially the sedimentary rocks in view of the fact that Ohio is covered with them. The casual field observer is much more likely to find different types of rocks to identify than many of the minerals to which so much space is given. Otherwise the book is extremely well done. It fills a great need in Earth Science, a field in which our science teachers are generally less well prepared. It gives these teachers simple aids and projects from which they can advance with confidence. The authors and the state survey are to be commended for providing this manual to aid our science teachers.

JOHN R. COASH

RESULTS OF AN EXAMINATION OF METAL SPECIMENS FROM AN EXCAVATION OF SHAMSHIR GHAR, AFGHANISTAN[1]

EARLE R. CALEY AND WALLACE H. DEEBEL
Department of Chemistry, The Ohio State University, Columbus

The specimens discussed in this paper all came from the excavation of the cave site of Shamshir Ghar, Afghanistan, conducted by Dr. Louis Dupree of Harvard University. This site is situated in the Province of Kandahar, at 31° 35' N latitude and 65°30' E longitude, and is about 15 miles WNW of Kandahar city.

According to the findings of Dr. Dupree, this site was not occupied by man until after the beginning of the Christian Era. Four cultural levels were identified, all belonging to pre-Mongol invasions.

Seven specimens, all more or less corroded, were submitted by Dr. Dupree for examination. In four of them sufficient metal was present to provide samples for quantitative analyses. The descriptions of these specimens and the results of our examination now follow.

No. 1. *Bronze Coin.* Diameter = 17–18 mm. Weight = 2.37 g. Date = IX-X Cent. A. D. This coin was covered with such a dense and uniform layer of green corrosion products that no trace of a design could be distinguished before cleaning. After electrolysis, parts of a crude design were visible. For analysis the surface layers were filed away, and two samples of fairly clean metal were obtained. The results of the analysis are shown in table 1.

These results show that this coin was composed of a lead-tin bronze of rather high lead content. Because of lack of analyses no comparison with respect to composition can be made between this coin and other coins of similar provenance. However, it is interesting that bronze of about the same composition was used for late Greek coins struck in eastern provinces of the Roman Empire. In all probability the silver in the alloy was introduced as an accidental impurity in the lead made from argentiferous galena. Though the proportion of nickel present as an accidental impurity falls within the normal range for an ancient bronze, the proportion of cobalt is unusually high and may be indicative of copper from a particular source. There is nothing remarkable about the proportions of iron and zinc. The low summations are due to the presence of undetermined oxygen and other non-metallic impurities present in the form of corrosion products within the samples of metal.

No. 2. *Fragment of Thin Bronze.* Length = 25 mm. Width = 15 mm. maximum. Thickness = ca. 1 mm. Weight = 2.00 g. Date = IX-X Cent. A. D. This appeared to be a segment of a disc, though the complete object could have had a different shape. Faint traces of an ornamental design were visible on the surface, which was covered with a thin layer of dirt and corrosion products. These were removed mechanically as far as practicable in preparing the two samples for analysis. The results of the analysis are shown in table 2.

These results show that this fragment was also composed of a lead-tin bronze, but one of lower tin content and even higher lead content than the metal of the coin. It is not surprising that the percentages of copper and lead in the two samples are not in agreement, for in bronze of such high lead content the lead

[1]Paper presented at the Sixty-fourth Annual Meeting of the Ohio Academy of Science, Delaware, Ohio, April, 1955.

THE OHIO JOURNAL OF SCIENCE 55(5): 311, September, 1955.

is often irregularly distributed through the alloy. On comparing the figures of table 2 with those of table 1 it will be seen that both bronzes contained the same impurities and that all except nickel were present in higher proportion in the metal of the fragment. The parallelism between the percentages of silver and lead in all samples and the near constancy of the ratios of these percentages, shown in table 3, is a clear indication that the silver was introduced as an impurity

TABLE 1

Analysis of bronze coin

Sample	Cu %	Sn %	Pb %	Ag %	Fe %	Ni %	Co %	Zn %	Total %
A	76.86	5.75	14.96	0.12	0.12	0.15	0.08	0.20	98.24
B	77.07	5.78	15.14	0.16	0.06	0.11	0.03	0.14	98 49

TABLE 2

Analysis of fragment of object composed of thin bronze

Sample	Cu %	Sn %	Pb %	Ag %	Fe %	Ni %	Co %	Zn %	Total %
A	67.22	3.36	25.53	0.20	0.49	0.11	0.13	0.66	97.70
B	64.54	3.29	28.29	0.22	0.52	0.11	0.12	0.74	97.83

Table 3

Ratio of Silver to lead in bronzes

Pb %	Ag %	Ag/Pb Ratio
14.96	0.12	0.008
15.14	0.16	0.011
25.53	0.20	0.008
28.29	0.22	0.008

along with the lead. The higher proportion of cobalt in the metal of this fragment as compared to the proportion of nickel is still more indicative of the use of copper ore from some particular source. As might have been expected, the summations of the results in table 2 are low because of the presence of corrosion products in the samples. In general, the analysis shows that the object from which this fragment came was composed of a kind of bronze suitable for ornaments but not for objects requiring hardness or strength.

No. 3. *Fragment of Pin or Clasp.* Length = 32 mm. Weight = 3.84 g. Date = ca. X Cent. A. D. This fragment appeared to be part of the shank of some sort of a pin or clasp. Mold marks on its sides indicated that this part of the original object had been formed by casting in a two-piece mold. The

fragment was coated with a thin, rather uniform layer of greyish-white corrosion products that contained stannic oxide and basic lead carbonate. The underlying metal was soft and white. Preliminary qualitative tests showed the presence of tin and lead as the major components of this metal. For quantitative analysis the surface layer of corrosion products was filed away, and two samples of rather clean metal were obtained. However, since the metal of this fragment had undergone extensive intergranular corrosion, it was not possible to obtain samples free from corrosion products. The results of the analysis are shown in table 4.

These results show that this fragment was composed of an alloy of tin and lead containing iron and zinc in small proportion as accidental impurities. This alloy is a kind of pewter in which the proportion of tin is close to twice that of lead. The closeness of the ratio seems to indicate that the alloy was not the result of the accidental or careless combination of the two component metals, but was the result of the deliberate combination of carefully measured amounts.

No. 4. *Broken Piece of Iron Pin or Shaft.* Length = 69 mm. Weight = 4.86 g. Date = VII-VIII Cent. A. D. This was a deeply rusted short length of straight rod that still contained some residual metal in its core as shown by tests with a magnet. On removal of the layers of rust mechanically a short, thin piece of metal was isolated. This iron was so soft that it could be easily filed,

TABLE 4

Analysis of fragment of pewter

Sample	Sn %	Pb %	Zn %	Fe %	Total %
A	63.87	32.64	0.08	0.03	96.62
B	63.64	32.89	0.07	0.02	96.62

and so ductile that it could be flattened out easily by hammering. After heating it to bright redness and plunging it into cold water it still could be easily filed and was still so ductile that it could be hammered out without any sign of fracture. This behavior is characteristic of iron containing a very low proportion of carbon. The metal of this object must have been, therefore, wrought iron or a very mild steel.

No. 5. *Broken Piece of Iron Pin or Shaft.* Length = 45 mm. Weight = 4.37 g. Date = VII-VIII Cent. A. D. This was similar to No. 4 in appearance, though somewhat thicker, and it also contained a short, thin core of metal. This iron was noticeably more resistant to the file than the metal of No. 4, and much less ductile on hammering. After heating it to bright redness and plunging it into cold water it was still more resistant to the file, and on hammering it broke into small pieces. This behavior is characteristic of iron containing enough carbon to be classed as a steel. In view of the difference in behavior before and after heat treatment, it must be concluded that the metal of the object was a tempered steel.

No. 6. *Broken Piece of Iron Arrowhead.* Weight = 2.11 g. Date = VII-VIII Cent. A. D. This was part of a bayonet-shaped arrowhead with three ribs. No residual metal could be detected by means of a magnet. The form of this fragment and the extent to which it had rusted clearly seemed to indicate that the arrowhead had been fashioned from either wrought iron or steel but not from cast iron.

No. 7. *Irregular Flat Piece of Cast Iron.* Thickness = 3-5 mm. Weight = 48.6 g. Date = X-XIII Cent. A. D. This piece of cast iron broke readily on being struck with a hammer and showed the bright crystalline structure characteristic of white cast iron. It also had the characteristic hardness of this variety of cast iron, for it could be neither drilled nor filed with tools of ordinary steel. The layer of rust on its surface was not more than a few tenths of a millimeter in thickness, which is not surprising in view of the known resistance of cast iron to corrosion under natural conditions. This specimen appeared to be a fragment from an iron plate, or possibly a piece from the botton of a dish or pan. The fact that its thickness tapered regularly from 5 mm. at one end to 3 mm. at the other was perhaps an indication of the latter possibility. One side was smooth and the other slightly rough, which may indicate that the original object was cast in an open mold. A few small blowholes were present in the metal under the rough surface, and this also may be an indication of the use of this technique. Except for these blowholes the casting was sound. Separate samples were taken for the determination of the usual constituents of cast iron. The results of these determinations are show in table 5.

TABLE 5

Analysis of fragment of cast iron

C	Si	P	S	Mn
%	%	%	%	%
4.25	0.13	0.11	trace	trace

These results fully confirm the previous observations indicating that this fragment was composed of white cast iron. The proportion of carbon is too high for steel, and is nearly the maximum that can occur in cast iron. Iron containing this particular proportion of combined carbon is the so-called eutectic alloy of iron and carbon, the one that melts or solidifies at a lower temperature than any other combination of iron and carbon. This minimum temperature is 1140°C, a temperature not difficult to reach even in a primitive furnace. The percentages of the other constituents are much lower than in modern cast iron. Iron of such high purity could have been produced by smelting a carefully selected, pure iron ore with charcoal in a blast furnace maintained at a relatively low temperature by the application of a regulated flow of unheated air. The higher percentages of impurities in modern cast iron arise from the use of unselected ore, coke as a fuel, and the higher temperature arising from the use of a blast of preheated air. From the standpoint of quality, the iron in this specimen is superior to modern white cast iron.

GENERAL CONCLUSIONS

A surprising variety of alloys and metallurgical techniques are represented by these few specimens. Such a variety of alloys and skills indicate an advanced stage in the art of metallurgy that could have been reached only on the basis of a long background of experience and practice.

NOTES ON THE GENUS *HAPLOPOGON* AND A NEW SPECIES (*DIPTERA: ASILIDAE*)

C. H. MARTIN
Oregon State College, Corvallis

Pritchard (1941) was the first to recognize the genus *Haplopogon* Engel in North America, the genotype *H. nudus* Engel being described from Turkestan in 1930. The male gonocoxite indicates that the three North American species described in the past are closely related among themselves and to the genotype species. (The writer agrees with Michener (1944) in considering the musculated claspers of most male insects as primary copulatory organs.) The gonocoxite of *H. nudus*, *latus* (Coquillett), *bullatus* (Bromley), and *erinus* Pritchard, is excavated mesally and most deeply on the distal third to a half so that the distal portion forms a more or less narrow strap of various shapes which curves mesally. The gonocoxite of *H. triangulatus* n. sp. is not excavated mesally and the tip curves laterad rather than mesad. In other respects the new species rather closely resembles the other species.

Engel (1930) illustrates a longitudinal row of several short hairs on the dorsal, apical portion of the third antennal segment. Our North American species possess similar hairs and in addition a long hair just anterior to the short hairs. Apparently this is a characteristic of North America species. Engel does not illustrate such a long hair.

North American Species of *Haplopogon*

1. Antennal style longer than third segment; axillary cell same degree of brown as adjacent areas; body brown pollinose (Texas)..... *bullatus* (Bromley)
   Antennal style equal to or shorter than third segment; border of axillary cell either milky hyaline or a lighter brown than along anal vein; body gray pollinose..... 2
2. Margins of mesonotal vittae sharp; scutellum light yellowish brown pollinose; bristle at tip of style black (Texas)..... *latus* (Coquillett)
   Margins of mesonotal vittae diffuse; scutellum gray pollinose; bristle at tip of style white or translucent..... 3
3. Antennal style and third segment of equal length; anterior cross-vein closer to anterior basal cross-vein than to middle of cell; axillary cell light brown along border and darker along anal vein (Arizona)..... *erinus* Pritchard
   Antennal style half as long as third segment; anterior cross-vein distad of middle of discal cell; axillary cell milky white hyaline except some light brown along anal vein (Texas)..... *triangulatus* n. sp.

Haplopogon latus (Coquillett)

Holopogon latus Coquillett, Jour. N. Y. Ent. Soc. 12: 33. 1904.
Holopogon lautus Back, Trans. Amer. Ent. Soc. 35: 314. 1909.
Holopogon lautus Bromley, Ann. Ent. Soc. Amer. 27: 98. 1934.
Haplopogon lautus Pritchard, Ann. Ent. Soc. Amer. 34: 351. 1941.

Authors have erroneously referred to this species as *H. lautus*. In the original description the spelling is *latus* and the writer found that the spelling on the type label at the U. S. National Museum is *latus*.

Notes: The type specimen may be a late teneral. Lateral margins of thoracic notum yellowish white pollinose behind the transverse suture; anterior to suture gray pollinose. Specimens at hand agree with this note. Dorsum of abdomen of type chocolate brown, covered with a thin gray pollinosity. One specimen at

hand has a chocolate brown area because of greasing, while on a second the entire dorsum of the abdomen is heavily gray pollinose.

Type specimen is a single male, Brownsville, Texas. Pritchard reports from Donna, Texas, March 3—April 7, 1935 (J. W. Monk). Specimens at hand from Corrizo Springs, Texas, April 14, 1949 (Michener-Beamer).

Haplopogon triangulatus, n. sp.

Male:—Length 4 mm. Head about 1.6 times longer than wide; black; white pollinose; vestiture white; mystax thin, confined to lower two-thirds of face; 10 ocellar bristles. Occiput thinly clothed with white bristles and hairs. Two proximal antennal segments subequal; third segment a little longer than combined length of proximal two; two-segmented style slightly less than half the length of the third segment, distal segment of style rather smooth with translucent spine at tip. Antennae blackish brown; row pale bristles on dorsal side of third segment, anterior bristle longer than style.

Thorax black; lateral margins gray pollinose, disc yellowish-brown pollinose, no distinct metanotal stripes, anteriorly on pronotum rubbed? black area; vestiture of thorax sparse, white; scutellum gray pollinose, sparsely clothed with pale hair. Legs black, vestiture white; claws yellow bases, black tips; white pulvilli.

Wings fulvus proximally; axillary cell fulvus along veins and diffusing into hyaline border; anal cell closed, petiolate, fulvus; fourth and fifth posterior cell lightly suffused fulvus at bases and hyaline in border; other posterior cells hyaline; milky white stippling over entire wing, stronger in hyaline areas. Distal knobs of halteres reddish-yellow brown.

Abdomen black, gray pollinose; sparsely clothed with pale hair longer laterally than dorsally. Genitalia black, gonocoxite (superior forceps of Pritchard) triangular in shape, tip curves ventrad and laterad.

Female:—Length 5 mm. Similar to male.

Holotype: Male, Davis Mts., Texas, June 9, 1954. D. J. and J. N. Knull, Colls.

Allotype: Female, same data.

Paratypes: two females and a male.

Holotype, allotype, and a paratype in collection of Ohio State University. Paratypes in writer's collection.

LITERATURE CITED

Engel, E. O. 1925–30. *In* Lintner, Die Fliegen der palaearktischen Region, 24, Asilidae, Stuttgart: 491 pp.

Pritchard, A. E. 1941. The genus *Haplopogon* in the new world, with the description of *erinus* n. sp. Ann. Ent. Soc. Amer. 34: 350–354.

Michener, C. D. 1944. A comparative study of the appendages of the eighth and ninth abdominal segments of insects. Ann. Ent. Soc. Amer. 37: 336–351.

Sex in Microorganisms. *D. H. Wenrich, I. F. Lewis,* and *J. R. Raper,* Editors. American Assoc. for the Advancement of Science, Washington. 1954. 362 pp. Illus. $5.75.

The book presents evaluations of the summarized literature dealing with the evidence for sexual reproduction in viruses, bacteria, fungi, unicellular algae, and protozoa. Chapters on the physiology of fertilization in ciliates and on the origin and evolution of sex concludes this symposium by recognized authorities. The reader is apt to find the latter chapter disappointingly superficial and almost completely lacking consideration of the botanist's points of view.

RICHARD A. POPHAM

NOTES ON THE LIFE CYCLES OF THREE PARASITES OF THE PITCH TWIG MOTH

WILLIAM E. MILLER
Ohio Agricultural Experiment Station, Wooster, Ohio

Four species of parasites, 3 primary and 1 secondary, have been reared by the writer from the pitch twig moth, *Petrova comstockiana* (Fernald) (Lepidoptera, Olethreutidae) in Ohio. In descending order of frequency of parasitization, the parasites are *Agathis pini* (Muesebeck), *Perilampus fulvicornis* Ashmead (secondary through *Agathis pini*), *Hyssopus thymus* Girault, and *Calliephialtes comstockii* (Cresson)[1]. *Agathis pini* was found to be a major influence in the natural control of the pitch twig moth in Ohio. This paper contains information on all parasites listed above except *Calliephialtes comstockii*. Methods of handling and observing parasites were the same as reported in a previous work (Miller, 1953).

The host develops in current growth twigs of various species of pine. It has a univoltine life cycle. The partly grown larva overwinters, and adults issue in May and June. A detailed account of the biology of the pitch twig moth is in preparation.

Agathis pini

The number of larval instars was not determined, but two general types of larvae were recognized: polypodeiform and fusiform. Polypodeiform (first instar) larvae were commonly found within young pitch twig moth caterpillars during the summer after July 1. Parasite larvae collected in July and August measured approximately 0.8 mm. in length. (Hosts had been killed in Peterson's K. A. A. D. killing mixture and preserved in 95 percent ethanol.) The polypodeiform larvae persisted in their hosts with only slight increase in size until the following spring. Meanwhile, the parasitized caterpillars proceeded normally in development and activity.

In the latter part of April, the parasite larvae began to grow rapidly. By April 23, 1952, two out of 8 larvae had transformed into fusiform individuals. Two weeks later the majority of larvae were fusiform and were from one-half to two-thirds the length of their hosts. The dissection records reveal that 100 percent of the twig moths found in the larval stage after May 1 had been parasitized by *A. pini*. Parasitized caterpillars never attained the pupal stage. They usually remained active until they completed the normal adult exit routes which later were used by the parasite adults. At maturity, beginning about May 10, *A. pini* larvae left the bodies of their hosts and spun cocoons in the host burrows. Nine days later only a few parasite larvae were still in their hosts. In an earlier paper (Miller, 1953), it was incorrectly stated that larvae of this species were external feeders. Cocoons prepared by the parasite larvae were cylindrical, white, and usually thick and opaque toward the cephalic end and thin and translucent toward the caudal end. Small portions of the cocoons were removed in the insectary so that the occupants could be observed. Several opened cocoons were completely repaired by the insects and had to be reopened for observation. Meconia were expelled in the cocoons from 2 to 4 days before pupation. Five larvae (2 males and 3 females) that were under observation transformed to pupae between May 20 and 27, 1952. The duration of their pupal periods varied from

[1]Braconidae, Chalcidoidea, and Ichneumonidae determined respectively by C. F. W. Muesebeck, B. D. Burks, and L. M. Walkley, of the Insect Identification and Parasite Introduction Section, U. S. Department of Agriculture, Washington, D. C.

THE OHIO JOURNAL OF SCIENCE 55(5): 317, September, 1955.

10 to 21 days and averaged 15 days. At every stage of development *A. pini* individuals were observed to be solitary.

In 1951 the single annual brood of adults emerged throughout June (Miller, 1953), and in 1952 they issued from June 10 through 19. On the initial dates of parasite emergence, host emergence was still underway in 1951, but complete in 1952.

Polypodeiform larvae never occupied any particular site within their hosts. However, during the latter part of larval development, the fusiform larvae were always situated in the caudal ends of hosts. Also, in 13 out of 15 instances, fusiform larvae were oriented with their heads in the same uniform direction as the heads of their hosts. This position of parasites and hosts corresponds with the location of the exits prepared by the hosts.

The short host list for *Agathis pini* (Muesebeck and Walkley, 1951) and the high frequency with which the parasite occurs on the pitch twig moth suggest that *A. pini* is highly host specific for this moth. The parasite has also been recorded from the pitch twig moth in New York and Maine (Muesebeck, 1940).

The 3-year insectary emergence record indicated that 40 percent of *A. pini* individuals had been parasitized by the hyperparasite *Perilampus fulvicornis*.

Perilampus fulvicornis

Three larval instars were distinguished by general appearance of developing larvae: first, the planidiform larva; second, the apoid larva; and third, the mature larva which is characterized by fleshy, segmental protuberances on the lateral aspect of the abdomen.

Dissections of over 100 twig moth larvae revealed 5 *Perilampus fulvicornis* planidia. One planidium was found in a caterpillar collected on July 18, 1952. This caterpillar apparently was not parasitized by *Agathis pini*. The remaining 4 planidia were solitary and were attached externally to the bodies of *A. pini* fusiform larvae removed from caterpillars collected in May, 1952. These planidia averaged 0.30 mm. in length. Due to their small size and inconspicuous coloration, many planidia may have been missed in the dissections.

A little later, larger planidia were discovered on 11 cocooned *A. pini* larvae that were developing under observation in the insectary. In two instances, two planidia were seen on the same host. At maturity, however, the hyperparasites were always solitary. There was never any indication that individuals completed development as primary parasites of the twig moth in the absence of parasitization by *A. pini*. The earliest transformation to the apoid larva took place in the insectary on May 25, 1952, and all individuals had attained this stage 4 days later. These transformations occurred from 5 to 10 days, or an average of 8 days, after expelling of meconia by host individuals. The immediate hosts (*A. pini*) did not live to pupation. The apoid period lasted from 4 to 6 days. The mature larvae were present 4 to 6 days prior to pupation. There was a 1 to 3-day prepupal period (Miller, 1953). Four individuals that reached the adult stage had pupal periods within the 8 to 13-day range noted in the earlier paper. In 1951, adults emerged over a 16-day period beginning on June 23 (Miller, 1953), and in 1952 over a 7-day period beginning June 18. When the first adults of *Perilampus fulvicornis* emerged, pitch twig moth emergence was complete, and *Agathis pini* emergence was nearly complete.

There is one hyperparasite generation annually, but most of the development of individuals takes place in less than one month. Approximately 10 months are spent in the first or planidial instar.

The species is recorded by Peck (1951) from many different hosts. It seems probable that there is a population within the species that maintains itself solely on the *Petrova comstockiana-Agathis pini* association.

Hyssopus thymus

The larvae are ectoparasitic and occur in aggregations of several individuals to a host. The winter is passed in the pupal stage within host burrows. In the insectary in 1953, the overwintering pupae produced adults from April 6 through 22. Adults of the next (summer) generation appeared about 6 weeks later, from May 25 through June 3. There is some evidence that at least a partial brood of adults emerged between the summer generation just mentioned and hibernation. Occasionally, adults and pupae were encountered in the field in August. The adults might have been long-lived members of the summer generation, but it seems improbable that pupae of the overwintering generation would occur this early in the season.

The number of individuals (pupae) per aggregation in 15 overwintering aggregations varied from 1 to 6 and averaged 2.5. The size of 6 summer aggregations varied from 1 to 19 pupae or mature larvae and averaged 10. Adults from two aggregations were sexed. One aggregation consisted of 7 females and 4 males, and the other consisted of 7 females and 1 male. From the time seven observed summer generation larvae expelled meconia to the time they pupated, a period of 1 to 4 days, averaging 3 days, elapsed. The pupal stage of 13 individuals of the summer generation ranged from 18 to 23 days and averaged 20 days in length. Previously, a shorter pupal period was reported for the species (Miller, 1953), but that observation was made on individuals from *Rhyacionia buoliana* (Schiffermüller) later in the season under warmer weather conditions. Pupae were always found to be oriented with their ventral surfaces upward. Adults from *Petrova comstockiana* were noted to be larger than those from *Rhyacionia buoliana.*

Hyssopus thymus appears capable of maintaining itself solely on *Petrova comstockiana.*

LITERATURE CITED

Miller, W. E. 1953. Biological notes on five hymenopterous parasites of pine bud and stem moths in Ohio. Ohio Jour. Sci. 53: 59–63.

Muesebeck, C. F. W. 1940. Two new reared species of *Bassus* (Hymenoptera:Braconidae). Entom. Soc. Wash. Proc. 42: 91–93.

———, and **L. M. Walkley.** 1951. Family Braconidae. In C. F. W. Muesebeck, K. V. Krombein, and H. K. Townes. Hymenoptera of America north of Mexico. (U. S.) Agric. Monogr. 2: 1–1420. p. 90–184.

Peck, O. 1951. Superfamily Chalcidoidea. In C. F. W. Muesebeck, K. V. Krombein, and H. K. Townes. Hymenoptera of America north of Mexico. (U.S.) Agric. Monogr. 2: 1–1420. p. 410–594.

ERRATA

In the article by Edward E. Terrell, Ohio Jour. Sci. 55: 1955, line 4 page 238, should read: *Heliopsis helianthoides* (L.) Sweet var. *helianthoides*. Ox-eye. Common along roadsides and in bottomlands; often with *Helianthus tuberosus*, (for in bottomlands; often with *Helianthus tuberosus*).

CONSULT OUR
Advertisers
☆
PATRONIZE OUR
Advertisers
☆
MENTION
THE
OHIO JOURNAL
of SCIENCE

NEW . . .
TURTOX BIOLOGY
Catalog No. 57
The new Turtox Catalog for the 1955 - 56 School year is now ready for mailing.
Have you received your free copy?
•
GENERAL BIOLOGICAL SUPPLY HOUSE, Inc.
8200 So. Hoyne Ave.
CHICAGO 20, ILLINOIS

Now Ready . . .

A History of Entomology
By HERBERT OSBORN

A brief History of Entomology from ancient times down to the present day. Dr. Osborn, dean of entomologists, puts in type a most interesting story of entomology through all its various divisions. 255 photographs with sketches of many of the prominent entomologists of all time.

A book for the amateur, the teacher, the student.

$4.50 postpaid

THE SPAHR & GLENN CO.
FIFTY EAST BROAD STREET, COLUMBUS 15, OHIO